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The relationship of bone density and physical activity, assessed by pedometry and accelerometry, in children

Powell, Sarah

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The relationship of bone density and physical activity, assessed by pedometry and accelerometry, in children.

Sarah Maley Powell

Thesis submitted for the Degree of Doctor of Philosophy of the University of Wales, Bangor.

School of Sport, Health and Exercise Sciences,

University of Wales, Bangor.

Spring, 2004.

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Acknowledgements

I would like to show my utmost gratitude to my first supervisor Dr. Ann Rowlands for her help, infinite wisdom, advice and support, in addition to her friendship. Also to my second supervisor Professor Roger Eston for his help guidance, support and friendship.

I would also like to thank Dr. David Ingledew for his help and guidance, Dr. Della Fazey for her support and encouragement, Dr. Dewi Jones for his help and enthusiasm with a particularly tricky area, Sarah Stevens for her assistance and humour, Dr. Vicky Marginson for cups of coffee and a big thank you to all the schools and participants for their involvement in my research.

I would like to thank the North Wales NHS Trust and North Wales Research Committee for funding studies one and five.

Finally, I would like to thank Luke, family and friends for their love, support and patience! You know who you are!

Declaration and Consent

I declare that this thesis has not been previously accepted in substance for any degree and is not being concurrently submitted in candidature for any degree. This thesis is the result of my own investigations.

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Summary

This thesis includes one qualitative literature review and five empirical studies examining aspects of a) the relationship between bone mass and physical activity in children aged 8-11 years using pedometry and accelerometry, and b) the variability and reliability of the RT3 accelerometer as a means of assessing physical activity patterns in children. Previous research has shown a positive relationship between physical activity and bone mineral density in children. However, study design appears to be confounded by the accuracy of measures of physical activity limiting conclusions that can be drawn. Prior to our investigation research investigating the relationship between bone mass and habitual physical activity measured by objective means in children is scarce, and no study had evaluated the technical performance of the RT3 accelerometer; a small lightweight triaxial accelerometer.

To examine the relationship between bone mass, physical activity and calcium intake methodology included; the objective assessment of habitual physical activity initially by pedometry and finally by accelerometry; dietary analysis of calcium intake; and dual energy X-ray absorptiometry measured bone area, mineral content and density of the whole body and proximal femur. To examine the variability and reliability of the RT3 accelerometer methodology included; the assessment of intraand inter-monitor variability by vibration assessment at increasing Hz levels; intermonitor reliability and variability by inducing a variety of human motions in a laboratory based environment; and epoch selection was assessed comparing the 60 to one second time sampling modes using children in a field setting.

The main findings were: a) steps per day, assessed by pedometry, explained a significant proportion of the variance in bone mass at the hip in children; b) the RT3 triaxial accelerometer was reliable across trials, although the anterioposterior vector recorded counts consistently higher than the mediolateral and vertical vectors; c) interunit variability of the RT3 was evident, particularly as activity intensity increased; d) use of the 60 second epoch setting may lead to inaccuracies when assessing activity of a vigorous intensity and above; and e) vigorous physical activity, assessed by the RT3 accelerometer, and calcium intake have an interactive effect on bone mass in children, whereby bone mineral content is only high when both vigorous activity and calcium intake are high.

In conclusion, this thesis has highlighted quality control procedures that need to be in place when using the RT3 accelerometer and presented evidence for a synergistic action of vigorous physical activity and calcium intake on bone mass in children.

Introduction to the Thesis

The accrual of bone mass during childhood and adolescence is a critical factor associated with the prevention of osteoporosis. It is therefore important to maximise bone mass during childhood in order to optimise the peak bone mass and offset subsequent loss. Although there is evidence for a positive relationship between habitual physical activity and bone mineral density in children, the research appears to be confounded by the accuracy of measures of physical activity. Indirect measures of physical activity including historical recall, diaries, questionnaires and observational surveys, have been widely used. Conceptually, the ideal solution for the assessment of physical activity is to use monitors that actually measure or track movement. However, research investigating the relationship between habitual physical activity measured by objective means and bone mass in children is scarce.

The RT3 accelerometer is a small and lightweight triaxial accelerometer that stores activity data for up to 21 days. It was introduced as a more easily usable device to replace the TriTrac-R3D. Prior to this thesis, no study had evaluated the technical performance of the RT3 accelerometer.

This thesis includes one qualitative literature review and five empirical studies examining various aspects of a) the relationship between bone mass and physical activity in children aged 8-11 years using pedometry and accelerometry, and b) the variability and reliability of the RT3 accelerometer as a means of assessing physical activity patterns in children.

Study one determined the relationship between habitual physical activity assessed by pedometry, calcium intake and bone mass in 8-11 year old boys and girls. The main findings were: a) physical activity explained a significant proportion of the variance in bone mineral content (BMC_R, residualised for bone area and body mass) at

the proximal femur and femoral neck; and b) calcium intake added to the variance explained at the proximal femur only. This study was presented at the Pediatric Work Physiology Conference in Corsendonk, Belgium (September, 2001) and published in Pediatric Exercise Science (14:358-68, 2002).

Study two determined the inter-monitor variability of a sample of 23 accelerometers, using an electronic vibrating jig. The main findings were: a) there were no differences in counts recorded on the X (vertical), Y (anterioposterior) and Z (mediolateral) axes at low acceleration (0.057g); b) the counts recorded along the Y axis were significantly higher than the counts at the X and Z axes at high accelerations (0.219g and 0.414g); and c) there were large coefficients of variation for both interand intra-instrument variability at low accelerations. This study was published in Medicine and Science in Sports and Exercise (35:1773-8, 2003).

Study three evaluated the reliability and variability of a sub-sample of eight RT3 accelerometers, across a variety of laboratory based physical activities. The main findings were: a) each individual monitor, along all axes, was reliable over trials; b) as intensity of activities increased the proportion of monitors that could differentiate between two adjacent activities decreased along all axes; and c) the variability between monitors within each activity increased along all axes as the intensity increased. This study was published in Medicine and Science in Sports and Exercise (36:324-30, 2004).

Study four evaluated the use of a 60- or 1-second epoch setting when using the RT3 accelerometer to assess habitual physical activity in children. The main finding was the overestimation of 'vigorous' activity and the underestimation of 'very hard' activity physical activity counts when using the 60-second epoch setting. This study has been submitted to Pediatric Exercise Science.

Study five examined the relationship between vigorous-intensity activity, calcium intake and bone mass as well as total physical activity and bone mass in children aged 8-11 years. Habitual physical activity was measured using accelerometry, and the main findings were: a) vigorous activity and calcium intake had an interactive effect on total body BMC (residualised for bone area and body mass) in both boys and girls and also at the hip in boys. The interaction demonstrated BMC was only high if both vigorous activity and calcium intake were high; and b) total physical activity explained variance in hip BMC in boys only. This study was presented at the Pediatric Work Physiology conference in Porto, Portugal (September 2003) and was awarded with the 'Young Investigators Award' in the poster presentation category. This study was published in the Journal of Applied Physiology (97:1203-8, 2004).

In conclusion, the use of pedometry and accelerometry provide further insights into the nature of the relationships between physical activity and bone mineral content in children. The findings support the biomedical perspective that high intensities of strain to the musculoskeletal system are more important to bone development, than the volume of activity. This thesis also provides evidence for the synergistic action of habitual vigorous physical activity and dietary calcium intake on bone mass in children.

The RT3 activity monitor allows intensity, frequency and duration of physical activity to be measured. The different time sampling intervals should be carefully considered when assessing vigorous intensity physical activity, as a more accurate picture of physical activity will be gained when using a shorter epoch setting. Intraand inter-monitor variability also exists. Therefore, it is recommended that all studies using the RT3 accelerometer perform trials to identify any outlying monitors and to assess the inter-monitor variability of RT3s over specific activities. This level of

quality control would ensure greater confidence in physical activity data, allowing relationships with health to be examined.

Summary of Publications and Presentations

Publications

Rowlands, A.V., Powell, S.M., Eston, R.G. and Ingledew, D.K. Relationship between bone mass and habitual physical activity and calcium intake in 8-11 year-old boys and girls. *Pediatric Exercise Science*, 14:358-68, 2002.

Powell, S.M., Jones, D.I. and Rowlands, A.V. Technical variability of the RT3 accelerometer. *Medicine and Science in Sports and Exercise*, 35:1773-8, 2003.

Powell, S.M. and Rowlands, A.V. Intermonitor variability of the RT3 accelerometer during typical physical activities. *Medicine and Science in Sports and Exercise*, 36:324-30, 2004.

Rowlands, A.V., Ingledew, D.K., Powell, S.M. and Eston, R.G. Interactive effects of habitual physical activity and calcium intake on bone density in boys and girls. *Journal of Applied Physiology*, 97:1203-8, 2004.

Abstracts

Powell, S.M., Rowlands, A.V. and Eston, R.G. Activity associated differences in bone mineral density of 8-11 year-old children. *Pediatric Exercise Science*, 13:262, 2001.

Powell, S.M., Rowlands, A.V. and Eston, R.G. The relationship between levels of habitual physical activity and body fat in 8-to-11-year-old children. *Journal of Sports Sciences*, 20:41, 2002.

Powell, S.M., Rowlands, A.V., Eston, R.G. and Ingledew, D.K. The relationship between 4-7 day accelerometry measures of physical activity, calcium intake and bone density in boys and girls, aged 8-11 years. *Portuguese Journal of Sports Sciences*, 3:151, 2003.

Presentations

Powell, S.M., Rowlands, A.V. and Eston, R.G. The relationship between level of habitual physical activity and body adiposity in 8-11 year old children. British Association of Sport and Exercise Sciences Conference (Newport, Wales, 5th September, 2001).

Powell, S.M., Rowlands, A.V. and Eston, R.G. Activity associated differences in bone mineral density of 8-11 year-old children. 21st symposium of the European group of Pediatric Work Physiology (Corsendonk, Belgium, 13th September, 2001).

Powell, S.M., Rowlands, A.V., Eston, R.G. and Ingledew, D.K. The relationship between 4-7 day accelerometry measures of physical activity, calcium intake and bone density in boys and girls, aged 8-11 years. 22nd symposium of the Pediatric Work Physiology group (Porto, Portugal, 16th September, 2003).

Awards

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Young Investigators Award at the 22nd symposium of the Pediatric Work Physiology group (Porto, Portugal, 18th September, 2003).

Chapter 1

Introduction

Osteoporosis literally means 'porous bones'. It is a disease characterised by low bone mass and micro-architectural deterioration of bone tissue, leading to increased bone fragility and a consequent increase in fracture risk (Drinkwater, 1994, Figure 1).

In the UK an estimated three million people suffer from osteoporosis, with one in three women and one in twelve men over the age of fifty having the disease. Osteoporosis costs the NHS and government over £1.7 billion each year, i.e. £5 million a day (National Osteoporosis Society, 2003). There is no cure for osteoporosis once the disease is established, and a large amount of bone will already have been lost by the time of fracture (Lysen and Walker, 1997). Currently, each year osteoporosis results in over 70,000 hip fractures and 40,000 spinal fractures (National Osteoporosis Society, 2003). Hip fractures cause the greatest number of deaths and lead to the most severe health problems and reduced quality of life (Hall et al., 2000).



Strong, dense trabecular bone.



Fragile, osteoporotic trabecular bone.

Figure 1. Healthy verses osteoporotic trabecular bone samples.

Bones are responsible for supporting loads placed upon the skeleton in an efficient and adaptable manner. Over the lifespan, bone change is dependent upon the unified work of bone cells; to resorb old cells and lay down new ones. Through these modelling and remodelling states a net increase in bone mass can be observed, as seen during growth, as can a net decrease, as seen with aging.

There is a peak bone mass [the highest bone mineral content during adulthood (Khan et al., 2001, p.111)] observed during the young adult years (Loucks, 1998; Bailey et al., 1999; Kemper, 2000) at which time an optimal amount of bone may offset future bone loss and delay the onset of osteoporosis (Lysen and Walker, 1997). As there is no cure for this disease once it is established, the accrual of a maximal amount of bone during childhood and adolescence is of utmost importance to skeletal integrity.

Higher bone mineral content (BMC) and density (BMD) have been observed in boys compared to girls during and after puberty, and also in the pre-pubertal years before sex hormones are released (Jones and Dwyer, 1998; Janz et al., 2001; Rowlands et al., 2002). Of the variance observed in bone mass, 60-80% can be explained by genetic influence (Bachrach, 2001), with the remaining 20-40% influenced by modifiable factors such as physical activity, calcium intake, smoking and oral contraceptive use (Rowlands et al., 2002; Janz et al., 2001; Khan et al., 2001, p.56).

Longitudinal intervention studies have reported a 4.6% increase (compared to controls) in site-specific BMC of seven year olds following 18 months of calcium supplementation (Lee et al., 1995) and a 9.3% increase (compared to controls) in site-specific BMC of pre-menarcheal females following nine months of aerobic and jump training (Heinonen et al., 2000). Whether these increases are maintained once the intervention ceases is under debate. Similarly, cross-sectional studies have shown

calcium intake to contribute between 1 and 10% of the variance in site-specific BMC (Barr et al., 2001; Rowlands et al., 2002) and habitual physical activity between 1.5 and 15% (Janz et al., 2001; Thorsen et al., 1999). Calcium appears necessary for physical activity to have an optimal bone stimulating action (Branca et al., 2001). Similarly reductions in benefits from calcium supplementation have been observed in the absence of mechanical loading (Parker, 1998). To date, calcium intake and physical activity have been considered or analysed as unrelated variables. However, there is a need to assess the interactive effects calcium intake and habitual physical activity may have on bone mass.

Although there is evidence for a positive relationship between physical activity and bone mass, questions may be raised as to the way in which physical activity has been measured and the effect the measurement method has on the detected size of the relationship. Indirect measures used to assess habitual physical activity include historical recall (Bass et al., 1998), diaries (Matkin et al., 1998), questionnaires (Bennell et al., 2000) and observational surveys (Cooper et al., 1995). However, Livingstone (1994) deemed self-report methods unreliable for use with children due to their relative lack of cognitive ability limiting recall of details concerning their activity patterns.

Conceptually, the ideal solution for activity assessment is the use of objective monitors that measure or track movement. Pedometers are motion counters which assess the amount of locomotion by total counting steps. Pedometer counts correlate highly with physical activity assessed by more sophisticated tools (Eston et al., 1998), and are useful to gauge an overall measure of physical activity in large populations (Rowlands, 2001). Uniaxial accelerometers are motion sensors which record the intensity, duration and frequency of vertical acceleration. They have been validated against oxygen consumption for use with children (Eston et al., 1998), and found to be

reliable (Trost et al., 1998) and practical tools for measuring habitual activity in the field (Janz et al., 1994). With the advent of triaxial accelerometers (e.g. TriTrac-R3D), a potentially more accurate measure of physical activity assessing multi-directional bodily movements is possible. The TriTrac-R3D has been validated against oxygen consumption with children in the laboratory (Eston et al., 1998) and in the field (Hendelman et al., 2000), and is reliable (Nichols et al., 1999). The main limitation with the TriTrac-R3D is its bulky size possibly limiting movement and maximising children's awareness of the monitor. The RT3 triaxial accelerometer was introduced as a smaller, lighter and user-friendlier alternative and seems ideal for use with children. The RT3 can record data at 1- or 60-second intervals (epochs) and to date it has been validated against oxygen consumption (Rowlands et al., 2004). However, no measures of its internal variability, reliability, placement or epoch effects have been investigated.

Structure of the Thesis

This thesis is structured as one qualitative literature review, three field-based empirical studies and two laboratory-based empirical studies. The literature review explores factors contributing to the relationship between calcium intake and physical activity with bone mass children, with special emphasis on the assessment of physical activity. In the first empirical study, the relationship calcium intake and physical activity have with bone mass in children is examined. Physical activity is assessed by pedometry and this is one of the first studies to consider the use of an objective activity assessment method when assessing its relationship with bone. However, it is not possible to consider intensity or duration of activity when using pedometry, therefore accelerometry is needed for the temporal assessment of activity intensity. To date no study has assessed the intra-or inter-monitor variability of the RT3

accelerometer under rigorous controlled testing or during standardised typical physical activities in the laboratory or in a free-living environment.

In the second study, the intra- and inter monitor variability of the RT3 accelerometer in a controlled electronic setting is examined. In the third study, the inter-monitor reliability and variability of the RT3 accelerometer is explored during various laboratory-based activities. In the fourth study, the RT3s epoch setting and placement effects during habitual free-living activity in children are examined. This provides a sound basis for a previously un-examined accelerometer to be used in empirical research. In the fifth and final empirical study, the RT3 accelerometer is used to clarify the potentially interactive relationship calcium intake and physical activity (total activity and vigorous activity) have with bone mass in children. Finally, a summary of the findings, conclusions and implications are presented.

Chapter 2

Literature Review

Bone

Bone is a unique tissue that is responsible for supporting loads imposed upon it. This apparently simple task demands that bones have enormous strength and



Figure 2. The structure of bone.

resilience while at the same time being lightweight and adaptable to ensure transportation is not a metabolic burden (Khan et al., 2001, p.3). Bone is made up from organic and inorganic compartments. The organic compartment contributes 25% to the weight of bone providing tensile strength. This is primarily made up from type I collagen and noncollagenous proteins (98%). It also includes bone cells (osteoblasts, osteocytes and osteoclasts 2%). The inorganic component, principally crystalline calcium

hydroxyapatite, contributes 70%

to the weight of bone, and provides resistance to compressive forces. The remaining 5% is water (Khan et al., 2001, p.3).

There are two types of bone: cortical and trabecular which account for 80 and 20% of the skeleton, respectively (Parker, 1998, Figure 2). Cortical bone forms the outer wall to all bones and is responsible for structure and protection. It is dense and calcified with 80-90% of its volume taken up by calcium. The bulk of cortical bone is found in the appendicular skeleton especially in the shafts of the long bones (Parfitt, 1988, p.47). Cortical bone has two types of surface. The endosteum is the internal surface and faces the bone marrow (Figure 3). Cells sited here are metabolically active and involved in bone formation and resorption. The periosteum is the external surface and aids appositional (cell wall thickness) growth (Khan et al., 2001, p.5). Trabecular bone is more structurally complex with a three-dimensional network housing either hematopoietic or fatty marrow (Parfitt, 1988, p.47). It has a greater metabolic rate than cortical bone and responds more quickly to changes in the environment (i.e. hormonal changes, Parfitt, 1988, p. 46). Trabecular bone is found mainly in the bones of the axial skeleton and the ends of long bones.

The mechanical competence of bone is a function of both its intrinsic material properties (mass, density, stiffness and strength) and its gross geometric characteristics (size, shape, cortical thickness, cross-sectional area and trabecular architecture) (Carter and Hayes, 1976). Bone mass is a determinant of its material properties. The distributions of bone mass and bone geometry are related to its strength and stiffness. Furthermore, the strength and stiffness of bone is a function of its density. Bone mineral content (BMC, g) refers to total grams of bone mineral as hydroxyapatite within a measured bone region. Bone mineral density (BMD, g.cm⁻²) is an areal measurement and refers to the grams of bone mineral per unit of bone area scanned. When bone samples are assessed in a laboratory setting under controlled loading conditions, BMD typically explains 60 to 80% of bone strength (Faulkner, 2000), and

small increases in BMD correlate with substantial increases in bone strength (Kiebzak and Miller, 2003).

Bone formation, resorption, modelling and remodelling

Bone is alive and constantly changing at a cellular level. There are specific bone cells (osteoblasts, osteocytes and osteoclasts), which regulate bone homeostasis by responding to various chemical and mechanical stimuli (Khan et al., 2001, p. 6).

Osteoblasts produce both collagen and ground matrix and are primarily involved in bone formation. They are found on the layer of bone they are producing and become calcified after a period of 10 days (Khan et al., 2001, p.7). Some active osteoblasts are incorporated into the bone matrix and buried beneath subsequent depositions. With time, these osteoblasts become either flat lining cells or osteocytes. Osteocytes are an important communication channel for mechanotransduction (the mechanism whereby physical activity influences bone cell function, Khan et al., 2001, p.11). They may also be involved in the repair of micro-fractures and may stabilise bone mineral by maintaining an appropriate environmental condition (Peck and Woods, 1988, p.10).

Osteoclasts have been recognized as the principal agents of bone resorption. Following their development mature osteoclasts migrate to bone surfaces where they attach and resorb a quantum of bone (Peck and Woods, 1988, p.2-6). They are large cells with a ruffled border. This border is the site of resorption, where its projections are intertwined with disaggregating bone mineral (Peck and Woods, 1988, p.2-6).

Osteoblasts and osteoclasts work in unison to form an organised bone cell activity enabling the process of bone growth and the adjustment of bone strength. This process is called bone modelling. Modelling improves bone strength by expanding the periosteal circumference and cortical thickness, for example, the change in bone shape and size during growth (Khan et al., 2001, p.16). During modelling either bone resorption or bone formation occur continuously on a surface for long periods of time without interruption resulting in a net gain of bone (Parfitt, 1988, p.51).

Bone remodelling serves the function of maintaining the biomechanical capability of the skeleton by preventing the accumulation of fatigue damage and safeguarding an adequate supply of young bone mineral to subserve mineral homeostasis (Parfitt, 1988, p.45). During remodelling, changes in the amount of bone are quite slow and changes in the shape of bones are barely perceptible. Bone remodelling is characterised by the cyclical erosion and repair of microscopic cavities with long periods of rest between cycles (Parfitt, 1988, p.51).

Bone over the life span: the peak bone mass

During growth, from conception until epiphyseal closure, bone formation exceeds resorption resulting in a net gain of bone, both trabecular from endochondral ossification and cortical from net periosteal apposition (Parfitt, 1988, p.46). As growth ceases there is a period of consolidation during which bone tissue density increases as bone turnover falls to a minimum (Parfitt, 1988, p.46). In a healthy adult skeleton an equal amount of bone is formed for that resorbed. However, in an ageing skeleton a shift towards resorption occurs producing an imbalance between formation and resorption. Consequently a net loss of bone is apparent (Figure 4).



Diagram 3. Internal bone structure.



Diagram 4. Net gain and loss of bone.

There is controversy concerning the specific time at which the peak of bone mass is achieved. Parfitt (1988), later supported by Loucks (1998), considered peak adult bone mass, representing the accumulated contributions of growth (90-95%) and consolidation (5-10%), to be reached between 35 and 40 years of age for cortical bone, possibly earlier for trabecular bone. Kemper (2000) however, regarded maximal bone mass to be reached in the late teens or early twenties. The Saskatchewan bone mineral accrual study spanning six years found 25% of the peak bone mass to be acquired in the two year period encircling peak height velocity, 12.5 years in girls and 14.1 years in boys, and at least 90% of peak bone mass accrued by 18 years of age (Bailey et al., 1999). Regardless of when the peak bone mass occurs it is approximately 25-30% higher in men than women and about 10% higher in people with African origins compared to Caucasians. However, within each demographic subgroup there are large differences among individuals with a coefficient of variation of approximately 15% (Parfitt, 1988, p.50).

Approximately 60-80% of the variance in peak bone mass has been attributed to heritability (Bachrach, 2001). Classic twin model studies have produced high estimates with family studies generally revealing lower estimates (Jones and Nguyen, 2000). Genes which may play a role in the control of bone mineral density include vitamin D receptor gene (VDR), the estrogen receptor (ER) gene, the apolipoprotein E4 gene, the interleukin genes, the parathyroid hormone receptor gene, and polymorphisms of the collagen gene (collagen IaISp1) (Khan et al., 2001, p.62).

To date it is unclear how much of the remaining 20-40% of possible variance in peak bone mass can be attributed to modifiable factors such as nutrition and physical activity.

Pubertal and pre-pubertal years

There is no cure for osteoporosis once the disease is established, and a large amount of bone will have already been lost by the time of fracture (Lysen and Walker, 1997). As at least 90% of total bone mass is accrued by the end of adolescence (Glastre et al., 1990; Bailey et al., 1999), the accrual of bone mass during childhood and adolescence and the optimisation of peak bone mass are critical factors to offset the impact of age-related bone loss and therefore, for the prevention or delay of osteoporosis.

Growth hormone (GH-IGF system) is responsible for growth during childhood (pre-pubertal years), whereas growth during puberty is also related to sex steroids, the concentrations of which vary with sexual maturation (Bass, 2000). Ratings of development were introduced by Tanner (1962) to assess pubertal status by means of sexual maturation. Five-stage scales (breast (female), genital (male) and pubic hair

(female and male) development) range from pre-pubertal to fully developed stages of sexual maturation. The concentration of hormones that influence bone: growth hormone, insulin-like growth factor one (IGF-1), oestrogen and androgens (male sex hormone), all increase during puberty (Khan et al., 2000). Specifically, growth hormone peaks during Tanner stages II-III in girls and IV-V in boys (Dunger et al., 1991) and serum oestrogen increases dramatically during Tanner stages III-IV (Khan et al., 2000). Tanner stage III corresponds to peak height velocity in adolescents (Bailey et al., 2000).

Theoretically, there should be no difference in the bone properties of boys and girls before puberty as these years are sex hormone independent. The disparity should be observed during puberty with the onset of sex hormones. This has been the reported case in several studies. Glastre et al. (1990) found no significant differences in BMD at the lumbar spine between pre-pubertal boys and girls. This is supported by Geusens et al. (1991), who reported no difference in BMC or BMD between pre-pubertal boys and girls at the radius, lumbar spine or total body. More recently, Jones and Dwyer (1998) found no significant differences at the lumbar spine of 8-year-old boys and girls; however, boys had a higher femoral neck BMD than the girls. Similarly, in a sample of 4-6 year olds, Janz et al. (2001) found BMC in boys to be significantly higher than girls at the lumbar spine, hip and total body with no correspondent differences in the total area of bone. Therefore, some studies indicate gender variance in bone mass before puberty; this may be explained by modifiable factors such as nutrition and habitual activity.

Nutrition

Calcium is an important nutrient for skeletal health throughout life, shown through the vast stores of calcium in the skeleton; 1400g in the mature male and

1200g in the mature female skeleton (Anderson, 2000). Although the cells of soft tissue have essential needs for calcium ions to support life, the focal need for calcium lies in skeletal utilization (Anderson, 2000).

Longitudinal calcium supplementation studies have provided a compelling link between calcium intake and bone accrual. Johnson et al. (1992) reported increased gains, compared to monozygotic twin controls, in spinal (2.8%) and radial (midshaft and distal, 5.1 and 3.8% respectively) BMD in male and female pre-pubertal subjects, when supplemented with 1000 mg, d^{-1} of calcium citrate over 36 months. Lee et al. (1995) reported similar gains in BMC at the lumbar spine (4.6%) and BMD at the radius (1.7%), compared to controls, in male and female 7 year-old subjects. This was despite only 300 mg.d⁻¹ of calcium carbonate being supplemented over only 18 months. Bonjour et al. (1997) also found increased gains, compared to controls, in radial (1.8%) and femoral (1.4%) BMD in 55 pre-pubertal girls who were supplemented for 12 months with 850 mg.d⁻¹ of calcium enriched foods. However, Bonjour et al., went on to report that the benefits of supplementation were 3.5-fold greater in the spontaneously low versus high calcium consumers, indicating there may be a threshold nutrient intake below which supplementation may be particularly beneficial to pre-pubertal children. They suggested a calcium intake below 800-900 mg.d⁻¹ might not be sufficient for optimal bone mass accrual in 7-9 year old prepubertal girls.

In a three year follow-up post supplementation, Bonjour et al. (2002) reported that the benefits to bone accrual were maintained in pre-pubertal girls, aged 7-9 years at baseline. Dibba et al. (2002) also found sustained gains in BMC at the midshaft radius 12- and 24-months after 12 months of supplementation ceased, in 160 children aged 8-12 years. In contrast, Slemenda et al. (1997) found that BMD gained by monozygotic twins aged 6-14 years (in comparison to twin controls) due to calcium

supplementation disappeared three years post withdrawal. In a follow-up to their earlier study (1995), Lee et al. (1996) also reported the increase in BMC due to calcium supplementation was not maintained 18 months after the intervention ceased. Subsequently, Lee et al. (1997) concluded that the effect of calcium supplementation on bone mineral gain may have reflected a temporary reduction in bone turnover rate.

The evidence has been provided for a causal link between supplemented calcium intake and bone mass, although whether or not increased gains in bone mineral can be maintained once supplementation is withdrawn is still questionable. However, the extent to which habitual calcium intake is related to bone mass is not clear. Cross-sectional studies have led to contradictory results. For example, positive correlations between BMD and calcium consumption have been reported in boys but not girls aged 4-20 years (Boot et al., 1997), girls aged 8-13 years (Ilich et al., 1998) and neither boys or girls at 7-11 or 16 years of age (VandenBergh et al., 1995; Sundberg et al., 2001). Barr et al. (2001) found calcium intake predicted baseline, two years post and two year change for BMC at the total body in 45 girls (mean age 10.6 \pm 0.6 years), explaining 1.6-5.3% of the variance. In support, Du et al. (2002) reported that milk intake accounted for 3.2% of the variance in BMC at the radius and ulna of 649 girls aged 12-14 years.

There are several methods available to assess habitual calcium intake. Barr et al. (2001) used a 'food frequency questionnaire' (FFQ), which was specifically designed to assess calcium intake and coupled it with three-day food diaries to produce a composite output measure of dietary calcium. The FFQ is not objective and can only estimate calcium intake based on the frequency of consumption of a selected list of foods ingested. However, it has previously been validated against three day dietary records (Ilich et al., 1998). Du et al. (2002) used a validated semi-quantitative FFQ to estimate milk consumption. The 103-item food list has been shown to

represent 86% of the calcium intake of Beijing residents (Du et al., 2002). Weighted food diaries, including both weekdays and weekend days, can offer habitual and quantitative reports of food ingested. Calcium intake can then be estimated using commercially available software. The general acceptability of this technique is apparent from its use as a criterion for validation studies (Ilich et al., 1998; Barrett-Connor, 1991). However, it must be acknowledged that weighted food diaries are subject to reporter-bias, under-reporting and increased food awareness whilst recording dietary intake.

Physical activity and bone

Julius Wolff described the ability of bone to adapt to the mechanical stresses placed upon it in the 19th century (Khan et al., 2001, p.13). Every physical activity generates a force either on specific bones or the whole skeleton. Bone cells respond to applied forces via the process of mechanotransduction (Khan et al., 2001, p.11). Within mechanotransduction there are four processes. Primarily, mechanical force is recognised by the osteocytes, or bone lining cells, and transformed to a form detectable by cells (mechanocoupling). Bone cells then attach to the collagen matrix (biomechanical coupling) and create a tension on the extra cellular matrix. This deformation stimulates osteoblastic activity (transmission of the biochemical signal), which produces new or rearranged bone (effector). Increased collagen and mineral apposition on the bone surface begins three to five days after a mechanical load is applied, and five to 12 days later the bone formation rate is increased largely due to the increased bone-forming surface (Khan et al., 2001, p.14-16). This results in either a net increase in bone, for example modelling in childhood, a re-energised bone surface, or a net decrease of bone, for example remodelling in later life.

The bone's response to a load being placed upon it will depend on the load's apparent strain magnitude, rate, number of cycles and distribution (Khan et al., 2001, p.27). Strain magnitude may be defined as the amount of relative change in bone length under mechanical loading, and strain rate as the rate at which strain develops and releases, which determines a bone's adaptive response (Khan et al., 2001, p.28). Based primarily on animal studies the link between strain magnitude, rate and number of cycles has been established. A mechanostat theory proposed by Frost (1987) has described a control system in which a minimum effective strain (MES) is necessary for bone maintenance (Figure 5).



Figure 5. Strain and bone's adaptive response.

Skeletal adaptations to mechanical usage: results from tibial loading studies in rats. Adapted from Bachrach (2001). In the trivial loading zone, minimal stimuli are provided to the bone and remodelling occurs. For example; over 17 weeks of bed rest, six healthy males lost up to 10.4% of site-specific bone mineral (Leblanc et al., 1990). Cosmonauts have also shown a 5.6% decrease in lumbar vertebral BMD after 14.5 months in space (Grigor'ev et al., 1998) and a 14% decrease in femoral neck bone mineral after 5.7 months in space (Oganov et al., 1992). In contrast, strains received in the physiological loading zone serve to provide enough stimuli to maintain a steady state of bone, which in turn maintains bone strength (Khan et al., 2001, p.28). When loading induced strain exceeds the modelling MES the overload zone is entered and bone modelling is stimulated to serve the mechanical demand.

It is most likely that strain produces fluid movement within the spaces surrounding the osteocytes creating direct stresses on the bone cells. This fluid may only be moved during cyclic loading. According to this theory, adaptation should only occur in response to dynamic loading, as static loads will not provide movement (Burr et al., 2002). Recent observations have shown static loading not only fails to provoke a response, but may suppress normal appositional growth. Robling et al. (2001_b) found static loading for 10 min.d⁻¹, over 14 days at high (17N) or low (8.5N) strain magnitude had no effect on endocortical formation rate, and suppressed periosteal formation in the growing male rat ulnae. In comparison, dynamic loading increased osteogenesis on both surfaces. Similarly, cross sectional studies support this notion, with swimmers (dynamic non-load bearing exercise) consistently being shown to have lower BMD than age-matched runners, gymnasts and jumpers (dynamic load-bearing activity). Furthermore, swimmers are shown to be no better off in terms of BMD than sedentary controls (Duncan et al., 2002; Taaffe et al., 2001; Courteix et al., 1998).

Strain rate is linearly proportional to strain magnitude and frequency (Burr et al., 2002). Turner et al. (1994) demonstrated that increasing the frequency of loading
(0.5 to 2.0 Hz) over 14 days, while maintaining a constant strain magnitude, elicited an increase in the bone formation of female rat tibiae. Later, Turner et al. (1995) created equivalent peak strains in adult rat tibiae however; the rates of strain were varied while the frequency and duration were kept constant. New bone formation was directly proportional to the rate of strain in the bone tissue. However, this may only be applicable to loading frequencies less than 2 Hz. Burr et al. (2002) commented that this is relevant as the majority of loading frequencies experienced by humans are below 2 Hz.

The osteogenic response of bone saturates rapidly in response to mechanical loading and bone cells require a recovery period to re-establish sensitivity (Burr et al., 2002). Umemura et al. (1997) assessed the bending rigidity of rat tibiae after eight weeks training of between five to 100 jumps.d⁻¹, for five days per week They found five jumps.d⁻¹ at a height of 40 cm were enough to create an increase in the cortical area of rat tibiae, and a few strain cycles were enough to saturate the bone response, as the tibiae showed no increased benefits from 100 compared to 10 jumps.d⁻¹. By allowing a recovery period between loading bouts, the effectiveness of the loading has been shown to be increased. Robling et al. (2002) found after 16 weeks of loading, increases in the BMC and areal density of rat ulnae were greatest when the loading was elicited in four blocks, separated by three hours recovery, compared to loading elicited in one continuous block. In previous work, Robling et al. (2001_a) stated eight hours was sufficient to re-establish complete mechanical sensitisation.

Strain distribution refers to the way strain is distributed across a section of bone (Khan et al., 2001, p.29). Bone cells maintain structural competence by adjusting bone architecture to remove or minimise variation from normal dynamic strain experienced (Rubin and Lanyon, 1985). Unusual strains of uneven distribution have been proposed to be more important for osteogenesis than frequency of cycles and

strain magnitudes arising from normal physical activities (Parker, 1998; Rubin and Lanyon, 1985).

Burr et al. (2002) reviewed the effects of biomechanical stress on bones in animals and concluded that activities involving high loading rates were most effective for increasing bone formation. Furthermore, when coupled with short periods of activity, with a four to eight hour rest period between bouts, an optimal osteogenic stimulus is created. Expanded periosteal circumference and cortical thickness with increased physical activity indicate that skeletal loading not only increases bone mineralization but also changes the bone shape (Specker and Binkley, 2003; Robling et al., 2001_b). Practical implications in humans for the optimisation of activity-induced osteogenesis, appear to be short, high loading, intermittent activities of unusual strain distributions with an appropriate recovery period (Parker, 1998). This is not accounted for in current health authority guidelines, where it is recommend that all young people participate in at least 60 minutes of moderate physical activity each day (Health Education Authority, 1998).

Ground reaction forces (GRF) are frequently used to investigate the effects of mechanical loading associated with weight-bearing activity in the human skeleton. Most commonly GRF is measured using a force plate as GRF is proportional to body mass and acceleration; the higher the impact of the activity the higher the GRF. In children, two-footed jumps from a height of 61 cm created GRFs which exceeded eight times body weight (Fuchs et al., 2001) and in adults jumping, running and aerobics induced GRFs three to six times body weight (Khan et al., 2001, p.30). Burdett (1982) conducted research concerning muscle groups and tendons operating on the ankle joint and concluded, with the addition of muscles and level arms, GRFs can reach 13 times body weight. This was recently supported by Giddings et al. (2000), who noted foot joint loads of four to five times body weight during walking

and eight to 11 times body weight during running. In older women (60-74 years), after 11 months of participating in an exercise regime designed to introduce stress to the skeleton through either GRFs (i.e., walking, jogging, stair climbing) or joint reaction forces (i.e., weight lifting, rowing), there were significant increases in BMD at the total body and lumbar spine (Kohrt et al., 1997).

Longitudinal studies of physical activity and bone mass

In humans, especially the young, only a few longitudinal studies have been performed. However, there is sufficient evidence that physical activity has a causal effect on bone mass. Morris et al. (1997) conducted a 10-month, high-impact exercise intervention consisting of aerobics, dancing, skipping, soccer and weight training for 30 minutes, three times a week, in 9-10 year old premenarcheal girls. Post intervention gains in BMC in the exercise group compared to controls were 5.5% greater at the total body, 5.5% greater at the lumbar spine, 7.3% greater at the proximal femur and 4.5% greater at the femoral neck. The exercise group significantly increased their lean mass and the changes in total body and regional lean mass emerged as the most robust determinant of BMC acquisition. Morris et al. concluded the increased rate of bone accrual may have been a response to the higher mechanical loading generated by the greater lean mass, supporting the strain and loading theories for bone accrual. A similar study by Bradney et al. (1998) conducted an eight-month, weight-bearing exercise intervention for 30 minutes, three times a week in 8-11 year old prepubertal boys. Post intervention gains in BMD in the exercise group compared to controls were 1.2% greater at the total body, 2.8% greater at the lumbar spine and 3.4% greater in the legs. Bradney et al. also measured bone geometry and hence estimated volumetric density; from this it was concluded that increases in femoral volumetric density may have been due to increases in cortical thickness.

More recently, McKay et al. (2000) conducted an eight-month, school-based jumping program consisting 10 tuck jumps three times per week with jumping, skipping and hopping which was incorporated into twice weekly exercise classes with pre- and early pubertal boys and girls. Post intervention BMD gains in the exercise group were 1.2% greater at the femoral trochanteric region of the proximal femur than in controls. Fuchs et al. (2001) took this further to explore the effects of a specific intensity, and hence force, generated upon bone mass. A seven-month intervention consisting of 100 two footed jumps from 61 cm boxes was performed three times per week, in pre-pubescent 5-10 year old boys and girls. Post intervention gains in BMC in the exercise group, compared to controls, were 4.5% greater at the femoral neck and 3.1% greater at the lumbar spine. Fuchs et al. concluded jumping from a height that causes ground reaction forces of eight times body weight is a safe, effective and simple method of improving bone mass at the hip and spine in children.

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In comparison, post-menarcheal adolescent girls do not receive the same gains in bone accrual from exercise interventions. Heinonen et al. (2000) compared the effects of a nine-month step aerobic intervention consisting of aerobic exercises and jumping training for 50 minutes, twice a week in 11-year-old pre-menarcheal and 13 year old post-menarcheal girls. Post intervention, the pre-menarcheal girls in the exercise group gained significantly more BMC than controls at the lumbar spine (8.6% compared to (cf.) 5.3%) and at the femoral neck (9.3% cf. 5.3%). However, the post-menarcheal girls showed no significant post-training increases in BMC when compared to the controls. Witzke and Snow (2000) also considered the effects of a nine-month plyometric jump training intervention consisting of hopping, jumping, bounding and box jumps but also weighted squats, lunges and calf raises for 30-45 minutes, three times per week in post-menarcheal girls (14.6 \pm 0.5 years of age). Although the exercise group showed improved knee extensor strength and

medial/lateral balance, there were no differences between the exercise and controls groups for BMC at the total body, femoral neck or lumbar spine. Similarly, over a 26week resistance training intervention in 16 year old postmenarcheal girls, significant increases in arm strength, knee flexion and squat press strength compared to controls was observed. Although, there were no greater increases in BMC in the exercising group than the control group (Blimkie et al., 1996).

The cellular and molecular basis for the differences in the response elicited by mature versus immature bone are not clear (Khan et al., p. 116). Intervention studies have begun the process of exploring the role of physical activity in bone mineral accrual. However, changes in a normalised pattern of physical activity and loading to a bone, i.e. due to an activity intervention programme, will subsequently change the functioning of the bone cell. The apparent effect this may have on the bone depends on the time of observation, and the long-term response may only be determined by waiting long enough for all remodelling transients to subside into a new steady-state (Parfitt, 1988, p.62). This length of time is so long, close to a year that slight changes in bone remodelling may occur with adequate frequency to ensure a steady state is never achieved (Parfitt, 1988, p.62). The resulting fluctuations in BMC will in most cases be small and this will weaken the significance of changes and make it more difficult to detect relationships in a group of subjects (Parfitt, 1988, p.62). To ensure the observation of an effect on bone due to an activity intervention programme, an adequate level and intensity of activity must be chosen coupled with regular monitoring of the bone surface, maturational status and growth over a lengthy intervention period. Given the complexity of these events, intervention studies are not only demanding but also difficult to design and carry out considering ethical limitations, time and resources. Therefore, it may be some time before the mechanisms behind the skeleton's response to loading are completely understood.

As discussed, few longitudinal studies have been performed and even fewer researchers have pursued follow-ups to identify whether or not bone accrual has been maintained once the stimulus ceases. However, there is evidence from athlete studies for the maintenance of increases in bone mass. In a four-year prospective follow-up study of male tennis players who started their playing career at the mean age of 11 years, Kontulainen et al. (1999) found previous exercise-induced bone gain in the playing arm was maintained 19 years later. This was 2.3 years post cessation of national level competition and despite decreased training frequency and load. This was later supported by Kontulainen et al. (2001) with a similar study in girls. In a prospective five-year follow-up of early (pre-menarche, mean age 10.5 years) and late (post-menarche, mean age 26 years) starting tennis and squash players, despite reduced training, exercise-induced bone gain was well maintained in both groups. However, at follow-up the bone gain was still higher in the early versus late starters (22% vs. 10%). This research group called for controlled interventions to confirm the findings that an exercise-induced bone gain can be well maintained with decreased activity. Later, Kontulainen et al. (2002) performed a 20-month follow-up study to assess whether BMC gained during a nine month jumping intervention was maintained. Regression analysis revealed the majority of variance in BMC gain was due to growth (height, weight and pubertal development). However, with the addition of intervention participation into the analysis, the exercise group were shown to have 4.9% higher BMC at the lumbar spine than controls 20-months later. A non-significant higher BMC (2-3%) at the proximal femur was also shown in the exercise group. Therefore, in the growing years some evidence exists to support maintained bone gain up to 20-months post training and, if the physical activity is maintained, albeit reduced, up to five years later.

Evidence has been provided for a causal link between imposed physical activity and bone mass. However, whether or not increased gains in bone mineral can be maintained once the exercise intervention ceases and the extent to which habitual physical activity, as opposed to structured training, plays a part in existing bone mass is not clear.

Sports participation and bone mass

Access to an activity intervention is not an option for many children. Therefore, it is important to understand whether sports participation plays a role in increasing bone mass.

Gymnastics is a high impact activity associated with around 102 and 217 impacts per session on the upper and lower body respectively, with peak magnitudes of four and 10 times body weight respectively (Daly et al., 1999). In both pre-pubertal and pre-menarcheal female gymnasts, BMD was significantly higher at the total body, lumbar spine and hip compared to controls (Courteix et al., 1998; Courteix et al., 1999; Nickols-Richardson et al., 2000). Racket sports also provide an excellence research base due to the impact-load being predominantly on one side of the body (i.e. dominant forearm). In pre-, peri- and post-pubertal female tennis players the playing arm had significantly higher BMD than the non-playing arm (mean difference ranging from two to 16%, Haapasalo et al., 1998). More recently Ducher et al. (2003) found a 19% difference in the BMC of the playing versus non-playing arm of 26 young tennis players (11.8 \pm 1.6 years) which was removed when lean tissue mass was taken into account, indicating the influence of high muscular activity and bone stress loading encountered in the playing arm. Greater BMD than age-matched controls has also been found in junior weightlifters (Conroy et al., 1993), elite female runners (Duncan

et al., 2002) and highly trained male judo and karate practitioners (Andreoli et al., 2001).

Witzke and Snow (2000) defined high-intensity activity as activities which generated forces greater than four times body weight, moderate-intensity as forces equivalent to between two and four times body weight, and low-intensity as forces equivalent to less than two times body weight. Grimston et al. (1993) provides evidence that activity intensities, which generate forces of three to 10 times body weight, stimulate bone accrual. They found children and adolescents aged 10-16 years, who engaged in impact-loading sports (running, gymnastics or dance), generating forces measured to be at least three and up to 10 times body weight, had significantly higher femoral neck BMD and a tendency for higher lumbar spine BMD than age, maturation, weight and training schedule matched children who engaged in activeload sports (swimming). However, this study lacks a comparison with sedentary controls. The effect the impact-loading sports had on BMD may have been underestimated by the comparison with elite swimmers who also engaged in weightbearing training, although significantly less than the impact-loading group.

The wealth of studies comparing sports participators with sedentary controls highlights the strong relationship physical training has with bone mass in adults (Suominen and Rahkila, 1991; Andreoli et al., 2001), adolescents (Duncan et al., 2002; Conroy et al., 1993) and children (Courteix et al., 1999; Nickols-Richardson et al., 2000). The extent of the relationship however, is still under debate and will depend on the intensity, duration and frequency of the activity.

Unfortunately, there is no bank of data regarding the minimum effective strain needed to stimulate osteogenesis at different ages throughout childhood and few studies have considered the ground reaction forces associated with activities their subjects are actively involved in. Therefore, a possibly useful measure of activity may

be the duration of high-intensity activities engaged in. Karlsson et al. (2001) assessed the impact of training duration in football players (mean age 23 years) on BMD. Three groups were devised; premier league players who trained for a mean 12 hrs.w⁻¹(hours per week), third-league players who trained a mean eight hrs.w⁻¹ and sixth-league players who trained a mean six hrs.w⁻¹. Training consisted of high-impact activities (running, kicking, jumping) involving unusual strain distributions (i.e. constant changes of direction). BMD was higher at the total body (6.8%), lumbar spine (13.2%) and femoral neck (12.7%) in the footballers when compared to age-matched sedentary controls. Femoral neck BMD was shown to increase by 3.3% for every hour trained from zero to six hrs.w⁻¹ and 0.7% for every extra hour trained above six hrs.w⁻¹. Scerpella et al. (2003) supported this research with pre-pubertal female gymnasts (7-11 years of age), where high (>eight hrs.w⁻¹) and low (one to eight hrs.w⁻¹) impact training groups were compared to controls. Analysis revealed the high training group to have consistently higher BMD than the low training group (2-5%) and both groups to have consistently higher BMD than controls (4-11%).

In both studies a duration-dependent relationship was shown between BMD and hours of impact training per week. Therefore research indicates even short durations (one to eight hrs.w⁻¹) of high-intensity activity may provide enough stimuli to pass the modeling minimum effective strain for bone accrual. The extent to which habitual physical activity as opposed to structured intervention or sport training may play a role in existing bone mass has yet to be clarified.

Habitual physical activity and bone mass

Slemenda et al. (1991) investigated the role of physical activity (assessed by questionnaire) in the bone mass of 112 children aged between five and 14 years. Positive significant correlations were found between total hours of weight-bearing

activities per week and BMD at the radius (r = 0.4), and hip (r = 0.34-0.41), which did not diminish after adjustments for age and gender. The data suggested children with activity levels one SD (2.7 hrs.d⁻¹) above the mean (1.6 hrs.d⁻¹) may have approximately 0.06 g.cm⁻² greater BMD, and may emerge from adolescence with 5-10% greater bone mass depending on the skeletal site. Boot et al. (1997) also found a significant positive correlation between physical activity (hours per week, assessed by questionnaire) and BMD at the total body and lumbar spine in 205 boys, aged between four and 20 years. However, possibly due to the low variance in the physical activity of girls, no association was found. In contrast, Illich et al. (1998) measured total energy expenditure in 456 preadolescent girls and found a positive association between bone mass (total body and radius) and energy expenditure. Thorsen et al. (1999) also investigated the relationship between habitual physical activity (hours per week) and BMD in adolescent boys (mean age 17 years). After body size and strength (quadriceps and hamstring isokinetic strength), hours per week of physical activity accounted for 15% of the variance in femoral neck BMD.

Janz et al. (2001) used a uniaxial accelerometer (CSA, model 7164) to objectively assess habitual physical activity in 368, 4-6 year old children. Since accelerometry allowed the intensity and duration of activity to be measured the relationship between not only total activity but also vigorous (>6 METs (metabolic equivalents)) activity with bone measures was investigated. Results revealed both activity measure was moderately correlated with hip and spinal BMC in boys and girls, and with total body BMC in boys only, after adjustments for age, mass and height. However at the hip, the site thought to be most influenced by weight-bearing activity and most sensitive to movement (Grimston et al., 1993; Thorsen et al., 1999; Janz et al., 2001), the correlations were slightly higher for vigorous activity (r = 0.25and 0.28 for boys and girls respectively) when compared to total activity (r = 0.20 and

0.25 for boys and girls respectively). Janz et al. went on to state combined physical activity measures explained 1.5-9.0% of the variance in bone measures (adjusted for age and body size), although vigorous activity was the most likely measure to enter the regression models. BMC was also 11.9% higher at the hip in children in the highest compared to the lowest quartile for vigorous activity. Although genetics, age, maturational development and body size are all important factors explaining much of the variance in bone mass, a proportion is explained by physical activity and calcium intake. However, it is not clear whether calcium intake or physical activity has the dominant effect on bone.

Interactive effect of physical activity and calcium intake.

It is documented that a substantial percentage of children and adolescents, especially females, do not consume enough dietary calcium, especially during the period of peak bone mass accretion (Barr, 1994; Weaver, 1996). Consequently, Anderson (2000) concluded as optimal amounts of calcium are consumed by relatively few, the beneficial effect of physical activity might dominate as a determinant of bone mass in early life. An adequate intake of calcium seems particularly important as calcium appears necessary for exercise to have an optimal bone stimulating action (Branca et al., 2001). Similarly, mechanical loading is necessary for calcium supplementation to be beneficial (Courteix et al., 2003). Therefore, both physical activity and calcium appear to need to be optimal for positive effects on bone. Molgaard et al. (2001) found a greater influence on bone mass from calcium intake than habitual physical activity, in 5-19 year old boys and girls. Size-adjusted BMC at the total body was significantly associated with average calcium intake in girls, but not boys. They noted, in a sample of relatively high calcium consumers (1000-12000 mg.d⁻¹), girls with double the mean calcium intake had 1.1% higher BMC. Over one

year in boys, the size-adjusted accretion of BMC was significantly associated with the change in calcium intake, and a doubling of calcium intake over the one year period was associated with 0.5% higher BMC accretion. Hours spent in high-intensity physical activity were not associated with BMC or BMC accretion in either boys or girls. To date, many studies have considered calcium intake and physical activity to work as separate entities. It is necessary to assess whether there is a synergistic action between calcium intake and physical activity. If this is shown to be the case it will be important to determine what combination of calcium and activity is optimal for bone accrual during the important years of growth.

Specker and Binkley (2003) examined the possible synergistic action of physical activity and calcium intake with a 12 month, randomised, placebo-controlled intervention trial of activity and calcium in 178 children aged 3-5 years. Children were assigned to either a fine or gross motor activities regime and sub-randomised to either calcium or placebo supplementation. The fine motor group performed 30 min.d⁻¹ of activities designed to keep them sitting quietly, in comparison the gross motor group performed 30 min.d⁻¹ of jumping, hopping and skipping activities (including five minutes to warm-up and cool-down). Calcium supplementation involved oral ingestion of 500 mg.d⁻¹ (five days a week) of calcium carbonate or placebo. After 12 months there was no difference in leg BMC gain between gross and fine motor groups receiving the placebo (38.2 ± 1.2 cf. 38.5 ± 1.3 g, respectively), compared to the 3.6g higher BMC increase shown in the gross verses fine motor group receiving the calcium supplement (40.9 ± 1.3 cf. 37.3 ± 1.4 g, respectively).

Significant interactions between activity and calcium groups were also found for the increase in cortical area (figure $6a^{1}$) and thickness (figure $6b^{1}$) of the tibia.

¹ Figures adapted from Specker and Binkley, 2003.

Among the calcium supplement group, the gain in both cortical area and thickness was greater in the gross compared to the fine motor group. However, results must be interpreted with caution as the fine motor group receiving the placebo supplementation showed similar gains.



Figure 6a¹. Increase in cortical area of Figure 6b¹. Increase in cortical thickness of the tibia by activity and supplement the tibia by activity and supplement groups.

Iuliano-Burns et al. (2003) found more consistent results in an 8.5-month, randomised, placebo-controlled intervention trial of activity and calcium in 66 preand early-pubertal girls. The girls were randomly assigned to one of four groups: lowimpact exercise with or without calcium fortified foods, or moderate-impact exercise with or without calcium fortified foods. The low-impact exercise group performed stretching and low-impact (GRF = one to two times body weight) dance routines for 20 minutes, three times a week. In comparison, the moderate-impact group performed hopping, jumping, and skipping based activities (GRF = one to four times body weight) for 20 minutes, three times a week, with progressive increases in intensity over the study period. Accounting for individual compliance, mean calcium intake \pm SD in the supplementation groups rose from 673 \pm 35 to 1121 \pm 45 mg.d⁻¹. There was little difference in femoral BMC gain between the moderate- and low-impact exercise groups receiving the placebo (25.4 ± 2.2 cf. 26.6 ± 2.1 g, respectively), whereas, the moderate-impact exercise group receiving the calcium fortified foods demonstrated a 10.5g higher BMC gain compared to the non-impact exercise group on non-calcium fortified foods group (31.7 ± 2.3 cf. 21.2 ± 2.6 g, respectively). A main effect of exercise was found at the tibia-fibula, with a 3% higher increase in BMC in the moderate- compared to low-impact exercise groups (15.6% cf. 12.7%, respectively, p < 0.05). A main effect for calcium was found at the radius-ulna, with a 4% higher increase in BMC in the calcium- compared to non-fortified food groups (p < 0.01).

These findings are supported by Courteix et al. (2003) who also found an interactive effect between activity and calcium on bone mass, however this time assessing habitual physical activity. One hundred and thirteen premenarcheal girls received either 800 mg.d⁻¹ calcium supplementation or placebo, and were subsequently split into habitually sedentary or weight bearing groups (1.2 ± 0.8 cf. 7.2 ± 4.0 hours per week). After 12 months of supplementation, the BMD gains (6.3%, p<0.05) at the total body of the weight-bearing activity with calcium group were significantly greater than all other three groups. Site-specific gains were also observed at the lumbar spine (p<0.05) and femoral neck (p<0.02). Twelve months following the intervention the weight bearing activity with calcium group maintained the significantly higher BMD values at the total body compared to the other three groups (p < 0.01). More research is needed to clarify the interactive relationship between physical activity and calcium intake on bone mass and whether the relationship exists when considering dietary calcium intake and habitual physical activity.

Activity Assessment

One major concern with research to date is the way in which physical activity has been assessed. The term 'physical activity' refers to "any bodily movement produced by skeletal muscles that results in energy expenditure" (Caspersen et al., 1985). However, in practice the definition of physical activity may differ depending on how it is measured and whether it includes measures of intensity, frequency and duration (Welk et al., 2000). Although there is evidence for a positive relationship between habitual physical activity and bone mineral density in children, the results appear to be confounded by the measures taken to assess physical activity. For example, Matkin et al. (1998) assessed the relationship between physical activity and bone mass using a three-day diary and a questionnaire based on the year's activities. They reported that the detected relationship varied in strength and direction depending on whether activity was assessed by the diary or questionnaire. Indirect measures of physical activity include historical recall (Bass et al., 1998; Molgaard et al., 2001), diaries (Matkin et al., 1998), questionnaires (Bennell et al., 2000; McKay et al., 2000; Kemper et al., 2000; Lehtonen-Veromaa et al., 2000) and observational surveys (Cooper et al., 1995).

Subjective measures

Self-report methods of activity assessment continue to be the most widely used type of physical activity measure (Sallis and Saelens, 2000) and provide a convenient way to assess large populations. However, there is widespread concern regarding the accuracy of self-report instruments (Welk et al., 2000).

Many self-report studies concentrate on sports participation rather than habitual daily activity (e.g. Lloyd et al., 2000; Jones and Dwyer, 1998). Even when studies focus on general physical activity, it is likely that sports activity is the most accurately reported due to the cognitive capacity needed to think abstractly and perform detailed recall of habitual physical activity (Welk et al., 2000). Sallis et al. (1993) observed good test-retest reliability for a seven-day recall interview (r = 0.77),

however, when compared to the criterion of heart rate monitoring, validity was only moderate (r = 0.53). Sallis et al. (1996) went on to validate a recall instrument under self-completion and personal interview situations, against heart rate monitoring and accelerometry, using fifth-grade American students. The self-administered recall correlated only moderately with heart rate (r = 0.57) and weakly with accelerometry (r = 0.30). Administering the questionnaire by interview did not improve the validity (r = 0.51 and r = 0.33 with heart rate and accelerometry respectively).

Overestimation of physical activity is common when using self-report methods. Sallis et al. (1996) found the average overestimation of moderate to vigorous activity was 48 minutes using the self-administered recall and 29 minutes using the interview-administered recall, when compared to accelerometry. Diaries are also subject to bias (Jakicic et al., 1998). Activity was overestimated, relative to accelerometer counts, by 45% of women using daily records. Self-report methods are sometimes aimed at the parents as children lack the cognitive ability to recall details concerning their activity patterns (Welk et al., 2000). However, over three days of physical activity assessment, only moderate correlations were found between TriTrac-R3D counts and a parent-reported questionnaire (r = 0.44) and a parent-reported diary (r = 0.42, O'Connor et al., 2003). Problems with limited response rates and instruction adherence (Sallis and Saelens, 2000) are also evident when using activity diaries. Due to these problems, Livingstone (1994) deemed self-report methods unreliable for use with children.

Objective measures

Bone mass is increased by dynamic activity involving high strains and unusual strain distributions (Parker, 1998; Turner and Robling, 2003). This type of activity may be particularly difficult to quantify using self-report. A meta-analysis on the

relationship between habitual physical activity and body fat in children highlighted that conclusions differed depending on whether the measure of activity was direct or indirect. Results revealed significantly different mean effect sizes for heart rate (r =0.00), questionnaire (r = -0.14), motion counters (r = -0.18) and observation (r = -0.39). The expected negative relationship was greatest for the observation method. However, there were no significant differences between the effect sizes observed when using direct observation and motion counters and hence, it was recommended that direct measures of movement be used to assess the relationships between activity level and health (Rowlands et al., 2000).

4

Conceptually, the ideal solution for the assessment of physical activity is the use of monitors that actually measure or track movement. The first motion counter or pedometer was designed roughly 500 years ago by Leonardo DaVinci (Freedson and Miller, 2000). The pedometer is typically placed at the waist and used to assess the amount of locomotion by counting steps. Vertical bodily acceleration triggers a lever arm to either rotate a ratchet or touch a digital pad to record one count. The pedometer's output measure is total counts, distance or calories and it does not provide any temporal data concerning activity patterns. It is not sensitive to any activity where the pedometer is not sited (i.e. isolated arm movement when the pedometer is sited at the waist or ankle) or static exercise, however, it is assumed that the contribution of static exercise to the total level of physical activity is negligible under normal daily living conditions (Bouten et al., 1997). Additionally, differences in spring tension can lead to high inter-unit and inter-model variability making comparisons between studies difficult (Freedson and Miller, 2000). Despite these problems the pedometer is ideal for population-based studies that require an objective, simple and inexpensive way to measure total habitual physical activity (Rowlands et al., 1997; Rowlands, 2001; Trost. 2001).

Pedometry correlates well with more sophisticated techniques such as triaxial accelerometry (Eston et al., 1998). Over a variety of treadmill and unregulated play activities, pedometers (worn at the hip) had a high correlation (r = 0.81) with oxygen uptake scaled for body size (VO₂, ml.kg^{-0.75}.min⁻¹), similar to that reported for heart rate (r = 0.79) and uniaxial accelerometry (r = 0.78) and slightly lower than triaxial accelerometry (r = 0.91). It was also noted that when only unregulated play activities were assessed, supposedly where the pedometer is least suited, the correlation with VO₂ was 0.92; significantly higher than corresponding correlations for the uniaxial accelerometer and heart rate with VO₂. In support, Hendelman et al. (2000) reported that when used as a tool to assess over ground walking, pedometer assessed steps were highly correlated with walking speed (r = 0.86) and VO₂ (r = 0.75). Additionally, Jones and Dwyer (1998) suggested that pedometry should be used when looking at the relationship between activity and BMD to facilitate accurate quantification of habitual physical activity and minimise measurement error.

In 1981, a portable motion sensor (Caltrac) with a piezo-electric accelerometer was developed to estimate energy expenditure during physical activity and commercially marketed (Muscle dynamics Fitness Network, Torrence, CA, USA). It could be attached to the waist and registered accelerations parallel to the vertical axis of the body. Reproducibility was very good (r = 0.94 over 14 activities, Montoye et al., 1983), and correlations with VO₂ and energy expenditure were very high (r = 0.76and 0.92 respectively) during four walking speeds in the laboratory (Balogun et al., 1989). In support Sallis et al. (1990) found moderate correlations with heart rate (r =0.42-0.54) and high correlations with oxygen consumption (r = 0.82) across two days of monitoring activity in the field. However, Haymes and Byrnes (1993) found the Caltrac generally overestimated energy expenditure during lab based locomotor activities above two mph. Conversely, during free-living activity the Caltrac

systematically underestimated the energy requirements when compared to indirect calorimetry (Shultz et al., 1988) and the doubly labelled water method (Gretebeck et al., 1991). The major limitation of the Caltrac is the final output. Total counts are accumulated for the whole sampling period, allowing no temporal analysis of activity patterns as with the pedometer (Freedson and Miller, 2000).

Modern accelerometers are superior to the Caltrac and pedometers as they can record accurate minute-by-minute accelerations and decelerations caused by bodily movements over several weeks. In this way, temporal analysis of movement data has become possible and permits analysis of total physical activity, time accumulated at different intensities of activity and the pattern of activity. The time resolution (epoch setting) of the accelerometer is important when assessing activity relevant to bone density as this allows short periods of intense activity to be captured. In children, the median duration of activities at any intensity level are reported to be less than 10 seconds and high intensity activities have a median duration of three seconds (Bailey et al., 1995). This is supported by Nilsson et al. (2002) who found a one minute epoch setting to be sufficient when measuring moderate intensity activity in children, however as activity intensity increased shorter (e.g. 10 seconds) epochs were needed to capture the time spent at a specific intensity. Accelerometers allow assessment using epoch settings between one and 60 seconds depending on the accelerometer. Other methods of assessing activity would likely miss these potentially important brief episodes of high intensity activity.

The Computer Science and Applications, Inc., (CSA, also known as the Actigraph, Shalimar, FL, USA) accelerometer is a small ($51 \times 38 \times 15$ mm), lightweight (43g) uniaxial accelerometer which records accelerations from 0.05 to two G's. Accelerations placed on the accelerometer act to create strain on a cantilever beam producing a proportional charge (Freedson and Miller, 2000). Each signal is

summed over a user-specified time interval and stored automatically resetting the monitor to zero. The CSA is initialised and downloaded via a computer interface allowing the user to set the start time and epoch.

In controlled, electronic laboratory tests the CSA has been shown to be a reliable tool, with an inter-instrument coefficient of variation (CV) of less than 5% and an intra-instrument CV of less than 2% (Metcalf et al., 2002). Laboratory testing has also revealed no significant difference between two CSA accelerometers worn simultaneously at each of three treadmill speeds (three, four and six km.h⁻¹) and a high inter-monitor reliability coefficient when averaged over the three speeds (r = 0.87, Trost et al., 1998). Counts were also highly correlated with energy expenditure (estimated from oxygen consumption), oxygen consumption (both r = 0.86-0.87), heart rate (r = 0.77) and treadmill speed (r = 0.9-0.89). Additionally, energy expenditure estimated by regression based on CSA counts was highly correlated with actual energy expenditure (r = 0.62-0.85). Eston et al. (1998) validated the CSA against oxygen consumption during typical children's activities (four, six, eight and 10 km.h⁻¹, catch, hopscotch and crayoning). Significant correlations were observed for all activities (r = 0.78) and unregulated play activities alone (r = 0.85) with VO₂ scaled for body size. In free-living situations, Janz et al. (1994) reported a high correlation between heart rate and CSA counts (r = 0.69), and the CSA exhibited higher between day stability than heart rate. Therefore, the CSA is a valid and reliable tool for use with children in the laboratory and free-living situations to gauge intensity, duration and frequency of physical activity. The main limitation of the CSA accelerometer is its uniaxial nature, possibly inhibiting the measurement of multi-directional bodily movements and hence predictions of energy expenditure.

This limitation of uniaxial accelerometry was noted early on by Ayen and Montoye (1988) who found energy expenditure during walking, running and squat

thrusts was better predicted by using three separate Caltracs mounted at right angles on the waist (r = 0.75) than by using a single vertical Caltrac (r = 0.65). This work was supported by Bouten et al. (1997) who found a triaxial accelerometer, composed of three orthogonally mounted uniaxial accelerometers, to have a high correlation (r =0.89) with energy expenditure over standardised laboratory activities.

The TriTrac-R3D (Hemokinetics, Inc., Madison, WI, USA) is a commercially available triaxial accelerometer. As with the CSA, the TriTrac-R3D recognises accelerations and decelerations from 0.05 to two G's, and is initialised and downloaded via a computer interface allowing the user to set the start time and sampling interval. A measure of intensity, duration and frequency is possible across three axes (vertical = X, anterioposterior = Y and mediolateral = Z) and a combined vector magnitude (VM= $(X^2 + Y^2 + Z^2)^{0.5}$).

The TriTrac-R3D has been successfully validated against energy expenditure measured by indirect calorimetry in the laboratory (mean r=0.86 over a range of lab based activities, Welk et al., 2000). Eston et al. (1998) reported the TriTrac-R3D to be a more accurate predictor of VO₂ than the CSA uniaxial accelerometer for treadmill activities (four, six, eight and 10 km.h⁻¹, r = 0.86 cf. 0.69), unregulated play activities (catch, hopscotch and crayoning, r = 0.93 cf. 0.85) and all activities combined (r = 0.89 cf. 0.78). These results were supported by Jakicic et al. (1999) and Nichols et al. (1999) who both reported significant correlations between TriTrac-R3D counts and energy expenditure measured by indirect calorimetry. Jakicic and colleagues, however, went on to report the TriTrac-R3D underestimates energy expenditure and the magnitude of this underestimation increases as exercise intensity increases (modes of exercise included; treadmill walking, running, stepping, cycling and slideboard). Conversely, Nichols and colleagues, reported significant overestimations of energy expenditure during treadmill walking although, high inter-monitor reliability between

hip placements (r = 0.73-0.87), intra-monitor reliability over time (r = 0.87-0.92) and significant differentiation between speeds (three, six and 10 km.h⁻¹) were found. It was noted however, the TriTrac-R3D did not differentiate between treadmill gradients (0-5%); as expected a change in gradient did not affect bodily accelerations or decelerations.

Similar results have been shown when assessing physical activity in the field. The TriTrac-R3D has been successfully validated against energy expenditure measured by indirect calorimetry in the field (r=0.62, Hendelman et al., 2000). Ott et al. (2000) reported TriTrac-R3D counts to be more highly correlated than the CSA, with predicted MET counts (r = 0.69 cf. 0.43) and heart rate (r = 0.73 cf. 0.64) during free-play activities (including throwing, catching, hopscotch and basketball) in children. In adults, Hendelman et al. (2000) assessed overland walking, golf, indoor and outdoor household tasks. TriTrac-R3D counts showed slightly higher correlations with estimated MET counts when compared with the CSA, for all activities (r = 0.62cf. 0.59 respectively) and walking only (r = 0.89 compared to 0.77). The main limitation with the TriTrac-R3D is its bulky size it measures $120 \times 65 \times 22$ mm and weighs 168g. This limits its practicality for use with children. The RT3 triaxial accelerometer is relatively small (71 × 56 × 28 mm) and light (65.2g), and replaced the TriTrac-R3D as a more researcher and user-friendly device (figure 7).



Figure 7. The RT3 and TriTrac-R3D accelerometers.

In our laboratory, the RT3 has been successfully validated against oxygen uptake in both children and adults, over a range of regulated and non-regulated activities (r = 0.87, Rowlands et al., 2004). To date no study has assessed the intra-or inter-monitor variability of the RT3 accelerometer under rigorous controlled testing or during standardised typical physical activities in the laboratory or in a free-living environment.

Therefore, it is important to a) to determine an appropriate objective measure of physical activity and confirm its intra- and inter-monitor variability and reliability, over a variety of activities in both laboratory and field settings, b) to investigate the relationship between habitual physical activity (assessed using objective measures), calcium intake and bone mass, and c) to clarify the interactive effect habitual physical activity, both total activity and quantity of high-intensity activity, may have with calcium intake on bone mass.

Chapter 3

Study 1

²Relationship between bone mass and habitual physical activity and calcium intake in 8-11 year old boys and girls

² This study was published in Pediatric Exercise Science: Rowlands, A.V., Powell, S.M., Eston, R.G. and Ingledew, D.K. Relationship between bone mass and habitual physical activity and calcium intake in 8-11-year-old boys and girls. *Ped. Exerc. Sci.*, 14:358-368, 2002.

This study was funded by the O. V. Jones bursary (£1500), North Wales NHS Trust (see appendix A1). The authors would like to thank the children, parents and headteachers form Ysgol Glan Cegin, Ysgol Hirael and Ysgol Y Borth, North Wales who volunteered to participate in this study.

Abstract

Purpose: This study aimed to determine the relationship between bone mineral content (BMC), habitual physical activity, and calcium intake in children. **Methods:** Fifty-seven children, aged 8-11 years, wore pedometers for seven days to assess habitual physical activity. Calcium intake was estimated by a four-day food diary. Bone mineral content (BMC) and areal density (BMD) were measured at the total proximal femur and femoral neck using dual energy X-ray absorptiometry. Regression analysis was used to assess contributions of physical activity and calcium intake to BMC, residualised for bone area and body mass. **Results:** Physical activity explained 11.6% of the variance in residualised BMC at the proximal femur and 14.3% at the femoral neck (p<0.05). Calcium intake added to the variance explained at the proximal femur only (9.8%, p<0.05). **Conclusion:** This study provides evidence for an association between BMC and habitual physical activity.

Introduction

There is no cure for osteoporosis once the disease is established, and a large amount of bone will have already been lost by the time of fracture (Bachrach, 2001). Lifetime risk of osteoporosis and related fractures depends on the peak bone mass achieved at skeletal maturity, and subsequent age-related bone loss (Cooper et al., 1995; Kemper, 2000). At least 90% of peak bone mass is accrued by the end of adolescence (Bachrach, 2001). Therefore, it is important to optimise early bone accrual.

Studies have shown that dancers and gymnasts have 5-15% higher bone mineral density (BMD) than controls, which highlights the importance of exercise for BMD (Bennell et al., 2000; Lehtonen-Veromaa et al., 2000). Retrospective analyses of retired gymnasts, tennis and squash players have provided evidence that increased BMD gained in childhood may be maintained into adult life (Bass et al., 1998; Konultainen et al., 2001).

Much research in this area is retrospective and/or limited to athletic populations. However, recent intervention studies have shown a causal relationship between physical activity and BMD, with gains in BMD demonstrated following seven to ten month intervention programmes in pre-pubertal boys (Bradney et al., 1998), pre-menarcheal girls (Heinonen et al., 2000; Morris et al., 1997), 6-10 year old children (Fuchs et al., 2001) and pre- and early pubescent children (McKay et al., 2000_b).

There is some evidence for a positive relationship between habitual physical activity and BMD in children (Bailey et al., 1999; Gunnes and Lehmann, 1996; Jones and Dwyer, 1998; Kemper, 2000; Kemper et al., 2000; Lloyd et al., 2000; McKay et al., 2000_a). However, research is confounded by indirect measurement of physical

activity, such as questionnaires (Bennell et al., 2000; Cooper et al., 1995; Kemper et al., 2000_b; Lehtonen-Veromaa et al., 2000; McKay et al., 2000). The type of self-report instrument used can also affect the reported relationship. Matkin et al. (1998) reported that the detected relationship between physical activity and bone mass varied in strength and direction, depending on whether activity was assessed by a three-day diary or an interview based on the year's activities.

Dynamic activity involving high strains and unusual strain distributions is considered to be the most osteogenic type of activity (Seeman, 1998). This may be particularly difficult to quantify using self-report. Self-report studies considering sports participation in addition to habitual daily activity have found sports participation to be more strongly related to BMD than habitual physical activity (Jones and Dwyer, 1998; Lloyd et al., 2000). Even when studies focus on general physical activity, it is likely that sports activity is the most accurately reported, as children are limited in their ability to recall details about their activity patterns. For this reason, Livingstone (1994) deemed self-report methods unreliable. A meta-analysis of the relationship between habitual physical activity and body fat in children reported significantly lower effect sizes from self-report instruments than from more objective measures of activity, e.g. pedometry (Rowlands et al., 2000).

Conceptually, the ideal solution for the assessment of physical activity is the use of monitors that directly measure movement. Pedometry correlates well with more sophisticated techniques such as triaxial accelerometry (Eston et al., 1998). Additionally, it is ideal for population-based studies that require an objective, simple and inexpensive way to measure habitual physical activity (Rowlands et al., 1997; Rowlands, 2001; Trost, 2001). Jones and Dwyer (1998) suggested that pedometry should be used when looking at the relationship between activity and BMD to

facilitate accurate quantification of habitual physical activity and minimise measurement error.

It is unclear whether physical activity or calcium intake is more important for bone mass accrual. Some research has shown a greater influence on bone mass from calcium supplementation than physical activity interventions (Ilick et al., 1998). Whilst others have shown the contrary (Ruiz et al., 1995; Welten et al., 1994). It is well documented that a substantial percentage of children and adolescents, especially females, do not consume and absorb enough dietary calcium during the period of peak bone mass accretion (Barr, 1994; Chan, 1991; Weaver, 1996). Anderson (2000) concluded that, since few children consume optimal amounts of calcium, the beneficial effect of physical activity might dominate as a determinant of bone mass in early life.

The aim of this study was to determine the relationship between bone mass, objectively measured physical activity, and calcium intake in 8-11 year old boys and girls.

Methods

Participants

Fifty-seven children were recruited from two primary schools in North Wales. Of these 57 participants, 29 were girls (mean \pm SD, age = 9.3 \pm 0.6 yrs, mass = 31.7 \pm 6.8 kg, height = 134.7 \pm 7.7 cm) and 28 were boys (age = 9.8 \pm 0.8 yrs, mass = 33.2 \pm 6.3 kg, height = 137.6 \pm 5.1 cm). All participants were healthy with no known diseases affecting bone metabolism.

Ethics approval was granted by the North Wales Health Authority Research Ethics Committee (see appendix A2). Written informed consent was obtained from all parents and verbal consent from all children (see appendix A3). Each participant and their parents were visited at home, where all procedures were verbally explained. A pack was administered containing: a pedometer with seven cable ties and written instructions; four food diary forms with written instructions; and a maturational status form for all girls.

Anthropometric and maturational assessment

Height was measured to the nearest 0.1cm and body mass to the nearest 0.1kg using a free-standing Seca stadiometer and Seca scales (Seca AG, Reinach, Switzerland).

Girls self-reported their pubertal status, using Tanner stages of breast development (Tanner, 1962) and whether they had begun menstruation. Pubertal status was not assessed in boys.

Calcium Intake

Parents were requested to aid their child in recording the time and weight of all food and drink consumed within four 24-hour periods, covering three weekdays and one weekend day. Diaries were analysed for average daily calcium content in milligrams using commercially available software (Dietmaster, Version 4, Swift Computer Systems Ltd., Surrey, UK).

Physical Activity

Physical activity was measured using pedometers (Yamax Digi-Walker, SW-200, Tokyo, Japan), sealed with cable ties so that the children could not read or tamper with the pedometers. Data were collected between the months September to November. Each child wore a pedometer for seven days, except for five children (mean = 6.9 ± 0.3 days, four children recording six days and one child recording five days). Therefore, each child met the recommendation of at least four to five days physical activity measurement (Trost et al., 2000). The pedometer was worn on the hip above the dominant leg, from the time the child got up in the morning until the time they went to bed. The parents recorded the times the pedometer was put on and taken off, and recorded the count at the end of each day, which involved unsealing and resealing the pedometer. Pedometer counts were not revealed to the child until the end of the week. The mean daily pedometer count was used as the output measure for physical activity.

Bone Mineral Content and Density

Bone area (cm²), bone mineral content (BMC, g) and areal bone mineral density (BMD, g.cm⁻²) were measured at the hip using dual energy X-ray absorptiometry (DXA, QDR-1500 Elite, Hologic Inc., Waltham, MA, USA, software version 7.10). The scans were analysed for the total proximal femur and femoral neck regions. Scans took place within one week of the completion of physical activity monitoring. The standard Hologic protocol for positioning was followed.

All scans were conducted and analysed by the same investigator in our laboratory. Quality assurance was performed daily by scanning a spine phantom supplied by the manufacturer. The in-vivo precision error of DXA in our laboratory, expressed as the coefficient of variation, is approximately 1.0% for the total proximal femur (see appendix B).

Statistical Analysis

Descriptive statistics were calculated for all variables. Independent *t*-tests were used to analyse gender differences. Levene's test was used to test for equality of

variances and, where violated, appropriate adjustments were made to the degrees of freedom.

Multiple regression equations were used to assess the relative contributions of physical activity and calcium intake to BMD at the proximal femur (BMD-PF) and femoral neck (BMD-FN), after accounting for body mass. Gender was subsequently added (female = 1, male = 2) to assess if any variance was explained over and above that explained by physical activity and calcium intake.

However, BMD is an areal density measurement (g.cm⁻²), which does not adequately account for differing body sizes (Prentice et al. 1994). To correct BMC for body size, regression analysis was used. The dependent variable was BMC (g), the independent variables were bone area (cm²) and body mass (kg). The residuals from this analysis were saved and labelled residualised BMC (BMC_R). Height was not included in the correction for body size because it did not account for any variance in BMC beyond that already accounted for by bone area and body mass (Parker, 1998). The correction for body size was carried out for both the total proximal femur (PF) and the femoral neck (FN). All further analyses were carried out on total proximal femur residualised BMC (BMC_R-PF) and femoral neck residualised BMC (BMC_R-FN).

The relative contributions of physical activity and calcium intake to BMC_R -PF and BMC_R -FN were assessed using multiple regression analysis. Gender was added as a third variable (female = 1, male = 2) to assess if any variance was explained over and above that explained by physical activity and calcium intake.

An alpha level of 0.05 was used for all statistical tests (see appendix C).

Results

Descriptive data, characterised by gender, are shown in Table 1. Boys significantly consumed more calcium and were more physically active than girls (p < 0.05, Table 1). Boys' BMD was 13.9% higher than girls' at the PF ($t_{55} = -5.2$, p < 0.01) and 11.7% higher at the FN ($t_{55} = -4.6$, p < 0.01).

Table 1. Descriptive statistics (mean \pm SD).

	Boys (n = 28)	Girls (n = 29)
Age (years)	9.8 ± 0.8**	9.3 ± 0.6
Mass (kg)	33.2 ± 6.3	31.7 ± 6.8
Height (cm)	137.5 ± 5.1	134.7 ± 7.7
Calcium intake (mg.d ⁻¹)	844.5 ± 322.5*	655.6 ± 217.6
Habitual physical activity	13551.5 ± 3993.1**	10430.4 ± 2526.9
(counts.d ⁻¹)		
Pubertal Stage	N/A	1.48 ± 0.5
BMC-PF (g)	14.853 ±3.265*	13.009 ± 2.752
BMD-PF (g.cm ⁻²)	0.745 ± 0.072**	0.654 ± 0.060
BMC-FN (g)	3.185 ± 0.490**	2.739 ± 0.390
BMD-FN (g.cm ⁻²)	0.688 ± 0.066**	0.616 ± 0.050

BMD = Bone mineral density, BMC = Bone mineral content, PF = Total proximal femur, FN = Femoral neck.

- * Boys significantly higher than girls, p < 0.05.
- ** Boys significantly higher than girls, p < 0.01.

The relative influences of physical activity, calcium intake and gender on BMD-PF and on BMD-FN are shown in Table 2. Physical activity explained 11.7 %

(p < 0.05) of the variance in BMD-PF. Calcium intake explained an additional 6.7 % (p < 0.05). Physical activity explained 12.2% (p < 0.05) of the variance in BMD-FN. Calcium intake did not add significantly to the variance explained.

Dependent	Predictor variables	Increment in R^2 when	Beta (standardised
variable	(in order of entry)	predictor variable	regression
		entered	coefficient) in final
			equation
BMD-PF	Physical Activity	0.117*	0.278*
	Calcium Intake	0.067*	0.267*
BMD-PF	Physical Activity	0.117*	0.132
	Calcium Intake	0.067*	0.165
	Gender	0.137*	0.421*
BMD-FN	Physical Activity	0.122*	0.299*
	Calcium Intake	0.042	0.212
BMD-FN	Physical Activity	0.122*	0.174
	Calcium Intake	0.042	0.125
	Gender	0.100*	0.361*

Table 2. Relative influence of lifestyle factors on BMD-PF and BMD-FN.

BMD-PF = total proximal femur BMD, BMD-FN = femoral neck BMD, Note: In all regression analyses body mass was entered as the first predictor variable, *p < 0.05.

The relative influences of physical activity, calcium intake and gender on BMC_R -PF and on BMC_R -FN are shown in Table 3. Physical activity explained 11.6% (p < 0.05) of the variance in BMC_R -PF. Calcium intake explained an additional 9.8% (p < 0.05). Physical activity explained 14.3% (p < 0.05) of the variance in BMC_R -FN. Calcium intake did not add significantly to the variance explained.

Dependent	Predictor variables	Increment in R^2 when	Beta (standardised
Variable	(in order of entry)	predictor variable	regression
		entered	coefficient) in final
			equation
BMC _R -PF	Physical Activity	0.116*	0.263*
	Calcium Intake	0.098*	0.323*
BMC _R -PF	Physical Activity	0.116*	0.101
	Calcium Intake	0.098*	0.197
	Gender	0.189*	0.490*
BMC _R -FN	Physical Activity	0.143*	0.333*
	Calcium Intake	0.034	0.189
BMC _R -FN	Physical Activity	0.143*	0.174
	Calcium Intake	0.034	0.066
	Gender	0.180*	0.478*

Table 3. Relative influence of lifestyle factors on BMC_R-PF and BMC_R-FN.

 BMC_R -PF = total proximal femur BMC residualised for bone area and body mass, BMC_R-FN = femoral neck BMC residualised for bone area and body mass, *p < 0.05.

The beta weights show that physical activity and calcium intake were significantly associated with BMD-PF and BMC_R-PF, when both lifestyle factors were considered (p < 0.05, Tables 2 and 3, respectively). However, only physical activity was significantly associated with BMD-FN and BMC_R-FN (p < 0.05, Tables 2 and 3, respectively).

Irrespective of the site, if gender was added as a predictor variable the beta weights for physical activity and calcium intake were no longer significant (Tables 2 and 3).

Discussion

This study used pedometry to assess the relationship between habitual physical activity and bone mass in children. Pedometry is an inexpensive, objective measure of physical activity (Rowlands et al., 1997) that compares well with more sophisticated accelerometry measures (Eston et al., 1998; Rowlands et al., 1999). It is possibly the only objective measure of activity ideal for population based studies. Habitual physical activity, assessed by steps per day, was significantly related to the size-adjusted bone mass and BMD of the total proximal femur and the femoral neck in 8 to 11 year old children. This relationship persisted after accounting for calcium intake.

Whether this data set was analysed using BMD or residualised BMC, as recommended by Prentice et al. (1994), did not affect the significance of the results. Therefore, the discussion will focus on the results from residualised BMC, as this measure avoids the assumptions necessary when calculating an areal density.

Habitual physical activity was a significant predictor of proximal femur BMC_R, explaining a total of 11.6% of the variance, and of femoral neck BMC_R, explaining 14.3% of the variance. This supports the findings of Thorsen et al. (1999) who identified the femoral neck as the site where the association between physical activity and BMD was the strongest in adolescent boys, with 15% of the variance explained. Thorsen et al. suggested that the load, or the forces experienced, during weight bearing activity were highest at the femoral neck, leading to the stronger relationships at this site. Unfortunately, no details of the assessment of physical activity were provided. Matkin et al. (1998) reported a positive relationship between hip BMD and activity, recorded in a three-day diary, in 9 - 25 year old males, though not females. In the same study, activity assessed by questionnaire was related to hip BMD in females. Matkin et al. concluded that poor correlations between the two self-

report instruments suggest that detection of the effect of physical activity on bone mass is instrument-dependent.

After controlling for physical activity, calcium intake was a significant predictor of proximal femur BMC_R, but not of femoral neck BMC_R. In an earlier study, Boot et al. (1997) reported calcium intake was a significant predictor of total body BMD in males aged 4-20 years, explaining 29.5% of the variance (p = 0.009), but not of lumbar spine BMD. BMD of the hip was not measured. No relationship between BMD and calcium intake was observed in girls. In support of this, Lloyd et al. (2000) found no correlation between calcium intake, bone gain and hip BMD, in 81 females aged 12 to 18 years.

In common with previous research (Boot et al., 1997; Gordon-Larsen et al., 1999; McKenzie et al., 2000; Rowlands et al., 1999) the boys' physical activity level was higher than that of the girls'. Calcium intake was also significantly higher in boys (by approximately 23%). Matkin et al. (1998) found a similar difference between the calcium intake of males and females aged between 9 and 25 years. Conversely, Boot et al. (1997) found no significant difference in the calcium intakes of 205 boys and 295 girls, aged 4 to 20 years. It could be speculated that any relationship identified between bone mass and these lifestyle factors could be due to boys having higher scores on habitual physical activity, calcium intake and bone mass (Jones and Dwyer, 1998; McKenzie et al., 2000). Indeed, in the current study, if gender was added as a predictor variable of size-adjusted BMC, physical activity and calcium intake were no longer significant predictors. This would indicate that gender might be a confounding variable leading to spurious correlations between physical activity and bone mass. We do not believe this to be the case for a number of reasons. First, the scatter plots in the current study show a trend for similar relationships in both genders. Second, relationships between formal activity or sports participation (assessed by

4 \$.
questionnaire) and bone mass have been reported independently for boys and girls (Jones and Dwyer, 1998; Matkin et al., 1998). Third, intervention studies have shown that formalised activity programmes can lead to an increase in bone mass in boys (Bradney et al. 1998), girls (Heinonen et al. 2000; Morris et al. 1997) and mixed groups (Fuchs et al., 2001; McKay et al., 2000_b). Ideally, to remove gender as a potential confounder, each gender needs to be analysed separately. However, the sample was not big enough to provide sufficient power to investigate the genders separately.

A further limitation of this study was that all children were not pre-pubertal. Fourteen of the 29 girls were Tanner stage 2, the remaining 15 being Tanner stage 1. The boys were not assessed for maturational status. However, no boy was above the age of 11, and research indicates that boys do not normally begin puberty until after 11 years of age (Albertsson-Wikland et al., 1994; Sizonenko, 1987). Based on this, we acknowledge the possibility that some of the boys may have reached Tanner stage 2, but it is highly unlikely that any had reached Tanner stage 3. Growth hormone secretion is largely responsible for growth before puberty (Bass, 2000), and the secretion rates do not differ by gender until Tanner stage 2 (Albertsson-Wikland et al., 1994). Additionally, Boot et al. (1997) studied 500 children and showed that the puberty-associated accumulation of bone mass started to increase at age 11 years in girls and 13 years in boys. In this study residualised BMC did not differ between Tanner stages (1 - 2) in girls (BMC_R-PF $t_{55} = 1.3$, p = 0.21, BMC_R-FN $t_{55} = 1.6$, p =0.11). Therefore, we are confident that pubertal status was not a confounder in this study. Nevertheless, we recommend that pubertal status is assessed in both boys and girls in future studies.

The sample of children was representative of British children of the same age for height and weight (Tanner and Whitehouse, 1984). The average girls' dietary

calcium intake was low compared to other studies (Heinonen et al., 2000; Jones and Dwyer, 1998; Matkin et al., 1998). Habitual physical activity scores (13551 ± 3993 counts.d⁻¹ for boys and 10430 ± 2527 counts.d⁻¹ for girls) were somewhat lower compared to scores previously reported for children from the same region (16035 ± 5998 counts.d⁻¹ for boys and 12728 ± 4026 counts.d⁻¹ for girls (Rowlands et al., 1999)), which may be due to the time of year of measurement. Bone mineral density values of the femoral neck (0.688 ± 0.066 g.cm⁻² in boys and 0.616 ± 0.050 g.cm⁻² in girls) were slightly higher than values reported for children of a similar age (0.66 ± 0.07 g.cm⁻² and 0.59 ± 0.07 g.cm⁻² in boys and girls, respectively (Jones and Dwyer, 1998)).

In conclusion, this study has provided evidence for an association between size-adjusted BMC (proximal femur and femoral neck) and habitual physical activity. This relationship persists after controlling for calcium intake. Additionally, calcium intake was independently related to size-adjusted BMC of the proximal femur. A strength of this study was the use of an objective measure of physical activity. This will have reduced the measurement error associated with the assessment of activity (Jones and Dwyer, 1998). However, pedometry provides only a measure of total activity. Accelerometry would also allow the intensity, frequency and duration of activity to be quantified. Assessment of the intensity of activity may be particularly important when investigating the relationship between physical activity and bone health.

Further research is warranted to explore the impact of physical activity and calcium intake on gender differences in bone mass in pre-pubertal children. To investigate this question it is important not only that objective tools are used to assess habitual physical activity, but also that boys' and girls' data are analysed separately.

Chapter 4

Study 2

³Technical variability of the RT3 accelerometer

³ This study was published in Medicine and Science in Sports and Exercise: Powell, S.M., Jones, D.I. and Rowlands, A.V. Technical variability of the RT3 accelerometer. *Med. Sci Sports Exerc.*, 35:1773-8, 2003.

Abstract

Purpose: The purpose of this study was to evaluate the technical performance of the RT3 triaxial accelerometer. Methods: Twenty-three RT3 accelerometers were subjected to a specific vibration along each sensitive axis in isolation, using a motorized vibration table which produced frequencies of 2.1, 5.1 and 10.2 Hz, respectively. Data were analyzed for frequency and axis effects and inter- and intrainstrument variability. Results: ANOVA showed a frequency by axis interaction ($F_{2.1,36.8}$ =19.9, p<0.001). Post hoc tests revealed the Y axis count to be significantly higher than the X and Z axes counts at 5.1 and 10.2 Hz. There was no difference in counts between axes at 2.1 Hz. Inter-instrument coefficients of variation (CV) decreased as frequency increased (21.9 to 26.7 % at 2.1 Hz, 6.3 to 9.0 % at 5.1 Hz and 4.2 to 7.2 % at 10.2 Hz). The intra class correlation (ICC) between RT3s was 0.99, regardless of the axis. Intra-instrument CV also decreased as frequency increased (2.1 to 56.2 %, 0.3 to 2.5 % and 0.2 to 2.9 % at 2.1, 5.1 and 10.2 Hz respectively. **Conclusions:** There were no differences in counts recorded on the X, Y and Z axes at 2.1 Hz, however the counts recorded along the Y axis were significantly higher than the counts at the X and Z axes at 5.1 and 10.2 Hz. Due to large coefficients of variation for both inter- and intra-instrument variability at 2.1 Hz, testing the interand intra-instrument variability of the accelerometers prior to use is recommended.

Introduction

Physical activity has long been recognized as a preventative tool for many chronic health conditions. In adults, both physical activity and physical fitness are inversely related to morbidity and mortality (Blair et al., 1989). However, quantification of the relationship may be hampered by the indirect measurement of physical activity, e.g. historical recall, diaries or questionnaires (Rowlands et al., 2000).

Conceptually, a possible solution for the assessment of physical activity is the use of monitors that directly measure movement (Rowlands, 2001). However, it must be recognized that activity monitors are unable to detect the metabolic cost associated with standing, upper body movements, static work, vertical lift and changes in gradient (Bassett, 2000). Accelerometers measure the accelerations of movement. A time sampling mechanism allows the capture of intensity, frequency and duration information. The Computer Science and Applications, Inc. activity monitor (CSA model 7164, also known as the actigraph or the WAM) is a small, lightweight uniaxial accelerometer, which is capable of storing activity data for up to 22 days. It has been validated against oxygen consumption during typical children's activities (Eston et al., 1998) and been shown to be a reliable tool, with an inter-instrument coefficient of variation (CV) of less than 5% and an intra-instrument CV of less than 2% (Metcalf et al., 2002).

The RT3 accelerometer is a small, lightweight triaxial accelerometer which stores activity data for up to 21 days. The three dimensional measure is potentially important when assessing activity. Eston et al. (1998) showed that a triaxial accelerometer (TriTrac-R3D) was a more accurate predictor of scaled oxygen uptake in children across a variety of activities than a uniaxial accelerometer. The TriTrac-

R3D has been successfully validated against energy expenditure measured by indirect calorimetry in the laboratory (mean r=0.86 over a range of lab based activities, Welk et al., 2000) and in the field (r=0.62, Hendleman et al., 2000). A limitation associated with the TriTrac-R3D is its bulky nature ($120 \times 65 \times 22$ mm, 168g). The RT3 is much smaller ($71 \times 56 \times 28$ mm, 65.2g) than the TriTrac-R3D and was introduced as a more researcher and user friendly device. To our knowledge no studies concerning the validity of the RT3 accelerometer have been published. However, the RT3 has been successfully validated against oxygen uptake in both children and adults, over a range of regulated and non-regulated activities in our own laboratory (r = 0.87, Rowlands et al., in press).

To date, no study has considered the inter-monitor technical variability of the RT3 accelerometer. Therefore, this study aims to determine the inter-monitor technical variability of a sample of RT3 accelerometers, on each orthogonal axis at three frequencies of motion.

Methods

Instrumentation and test procedures.

The RT3 accelerometer.

The RT3 (Stayhealthy, Inc., Monvovia, CA, USA) is a small ($71 \times 56 \times 28$ mm), lightweight (65.2g), battery-powered instrument used as an experimental tool for measuring the physical activity of people. It is worn clipped to the waistband as an 'accessory' during waking hours. Depending on its mode of operation, it can record data for up to 21 days, which is then downloaded to a PC for display and statistical processing. The sensor in the RT3 is an accelerometer sensitive along three orthogonal axes (X, Y and Z) which represent vertical, anterioposterior and

mediolateral motion, respectively. The acceleration is measured periodically, converted to a digital representation and processed to obtain an 'activity count' which is stored in memory. The exact relationship of the activity count to the acceleration (measured in m.s⁻² or g, where 1g = 9.81m.s⁻²) is not clear.

The RT3 has four modes of operation: mode one samples and stores activity counts on individual axes at 1s epochs; mode two samples and stores vector magnitude (a measure combining all three axes of motion) activity counts at 1s epochs; mode three samples and stores accumulated activity counts on individual axes over 1min epochs; and mode four samples and stores accumulated vector magnitude activity counts over 1min epochs. The latter two modes store less detail about activity but are more economical in their use of memory, allowing longer duration experiments to be performed. Epoch duration of one minute is generally used in the field and so was chosen for the trials reported. In this study the RT3 activity monitor was tested along each orthogonal axis separately, therefore whilst any one of the vectors was being tested the other two vectors should have recorded zero. As vector magnitude is a culmination of the three vectors it was not tested in this research. Twenty-three RT3 accelerometers were tested in total, all six months old and previously used in the field.

Vibration Table.

Each RT3 in turn was mounted securely in a test jig, which was screwed directly to the vibration table (Ling Dynamics 403). The vibration table was driven by a moving coil armature via a power amplifier. The amplifier input was provided by a signal generator, which was programmed accurately and reliably to vibrate the RT3 at frequencies of 2.1, 5.1 and 10.2 Hz.

There are two standard forms of industrial vibration tests, sinusoidal and random. Swept-frequency sinusoidal testing is easy to implement and usually used when the equipment being tested is subject to pulsating or oscillating forces of a periodic nature. Random testing is useful when the equipment may be subject to vibration of a stochastic nature, possibly exciting several resonant modes simultaneously. In this case, there was an advantage to the sinusoidal option. It is known the accelerometers used in the RT3 have a dynamic range of 0.05-2 g, are sensitive in the range 2-10 Hz and are calibrated at 5.3 Hz (personal communication, Stayhealthy 2002). The output of the RT3 accelerometer is not available directly to the user, neither is it possible or desirable to seek these signals by breaching the encapsulation. The only output available in practice is the RT3 activity count, the fastest sampling rate being 1 Hz. According to Shannon's sampling theorem (Franklin et al., 2002), the highest frequency component of the measured variable should therefore be 0.5 Hz, if it is to be recoverable from the samples. Using the well-known relationship between the acceleration and amplitude of sinusoidal motion at a known frequency, it becomes clear that achieving 0.05 g (at the lower end of the accelerometer's dynamic range) requires an amplitude of 49.7 mm at 0.5 Hz. This is outside the range of the Ling Dynamics 403, which has a maximum amplitude of 8.8 mm.

Therefore, the approach adopted was to vibrate the RT3 at frequencies of 2.1, 5.1 and 10.2 Hz which are slightly offset from a multiple of the 1 Hz sampling frequency. This creates low-frequency aliases of the true vibration frequencies which appear to have high peak accelerations at low displacements. The exact values of the accelerations are not important as long as they fall within the dynamic range of the RT3 accelerometer and are repeatable. A Kyowa AS-10B strain-gauge accelerometer (calibrated at 88.3 mV/g \pm 5 %) was mounted on the same plate as the RT3 test jig.

The sinusoidal waveform traces produced at the three frequencies and amplitudes (3.19mm, 2.09mm and 0.99mm, respectively) selected show that the peak test accelerations were 0.057g, 0.219g and 0.414g, respectively. When the RT3 counts are plotted against acceleration there is a linear relationship (Figure 8).



Figure 8. Variation of the RT3 count with peak acceleration (mean \pm SD).

Sampling Issues.

In practice the RT3 must be switched on at some random time relative to the vibration input. It is of interest to know whether this affects the measurements.

Considering sample values are given by:

$$y_s(kT) = sin(\omega_a kT + \phi)$$

where: $k = 0, 1, 2, 3 \dots 9$, ω_a = frequency of the alias, T = sample period, ϕ = phase difference between vibration waveform and sample times, $-\pi < \phi < \pi$.



Figure 9. Variation of phase with differing sample times.

Procedures adopted mean that irrespective of the time the RT3 is switched on an error of less than 5% in the output value is apparent.

Plotting $\omega_a = 0.1$ Hz, T = 1s and k = 0, 1, 2...3 (the remaining curves are similar) shows how the values measured at individual sample times vary with the phase, ϕ (Figure 9). It is only when $\phi = \frac{\pi}{10}, \frac{3\pi}{10}$ that the maximum value of 1 is recorded. However, even for the worst-case ϕ , a maximum value of 0.951 is recorded

– an error of less than 5%. So the method records a sample very near to the peak acceleration applied irrespective of the time that the RT3 is actually switched on.

Procedures.

Prior to each testing session, the apparatus were run for eight minutes at each of the three frequencies, to ensure that the mechanics were sufficiently warmed up. Each RT3, in turn, was placed in the test jig aligned so that vibration would occur along the X axis (Figure 10). Vibration of the test jig commenced at a frequency of 2.1 Hz. The frequency automatically increased to 5.1 Hz and subsequently to 10.2 Hz. Each frequency was maintained for eight minutes. The initial and final minute of data were discarded, leaving six complete epochs for analysis at each frequency. After each trial the test jig was removed, rotated and reaffixed to provide an alternative axis for vibration (Y and Z, respectively) and the procedures were repeated (Figures 11 and 12).



Figure 10. Test jig vibrating along the X axis: the arrow indicates the direction of movement of the vibration table.



Figure 11. Test jig vibrating along the Y axis: the arrow indicates the direction of movement of the vibration table.



Figure 12. Test jig vibrating along the Z axis: the arrow indicates the direction of movement of the vibration table.

Statistical Analysis.

Data was downloaded into Excel and imported into SPSS 9.0 for analysis. Descriptive data were calculated for all variables. *A priori* inclusion criteria defined outliers as activity counts greater than 2 standard deviations above or below the mean.

Frequency and axis effects.

A two way fully repeated measures ANOVA was used, with repeated measures on axis (X, Y and Z) and frequency (2.1, 5.1 and 10.2 Hz). Where the assumptions of sphericity were violated the Greenhouse Geisser correction factor was employed. Significant results were followed up using an adapted Tukey's test for repeated measures (Stevens, 1996).

Inter-instrument variation.

The relationship between accelerometers for each axis and for all axes combined was investigated using intra-class correlation coefficients (ICC) with a twoway random model for absolute agreement. The inter-instrument coefficient of variation (CV= SD/mean) was calculated for each axis at each frequency.

Intra-instrument variation.

The intra-instrument CV was calculated for each RT3, over the six epochs available for each axis at each frequency.

Alpha was set at p<0.05.

Results

Four RT3 accelerometers were identifies as outliers (two SDs above or below the mean). Data were analyzed including and excluding the outliers. Overall descriptive data for the total sample of 23 RT3's and the remaining 19 RT3s are presented in Table 4.

	2.1 Hz		5.1 H	z	10.2 Hz		
	N = 23	<i>N</i> = 19	<i>N</i> = 23	N = 19	N = 23	N = 19	
X	100.2	98.9	861.5	853.6	1707.4	1683.4	
	± 22.6	± 21.7	± 53.9	± 54.0	± 94.2	± 71.3	
Y	99.3	103.2	893.8*	912.6*	1815.8*	1847.9*	
	± 33.0	± 27.6	± 104.2	± 76.6	± 193.5	± 132.2	
Z	89.6	98.0	792.6**	847.1	1583.8†	1691.9	
	± 34.5	± 21.8	± 192.6	± 76.3	± 375.0	± 119.7	

Table 4. Descriptive statistics for each axis at each frequency, including all 23 monitors and excluding the four outliers (activity counts, mean \pm SD).

* = Y axis significantly higher than the X and Z axes, p<.05. $\dagger = Z$ axis significantly lower than the X axis, p<.05.

Frequency and axis effects.

Main effects for axis ($F_{1.1,19.4}=10577.7$, p <0.001) and frequency ($F_{1.5,26.1}=15.6$, p<0.001) were observed, accompanied by an interaction between the two ($F_{2.1,36.8}=19.9$, p<0.001). Post hoc tests revealed no significant difference between the axes at 2.1 Hz. However, the Y axis counts were significantly higher than the X and Z axes at 5.1 and 10.2 Hz (Figure 13). Inclusion of the outliers in the analysis still resulted in a frequency by axis interaction ($F_{1.4,31.1}=5.2$, p<0.02), with no significant difference in activity counts at 2.1 Hz. At 5.1 Hz and 10.2 Hz the Y axis was still significantly higher than the X and Z axes; however, the Z axis was also significantly lower than the Y and X axes.



Figure 13. Activity counts by axis and frequency.

* = Y axis significantly higher than the X and Z axes, p < 0.05.

Inter-Instrument variability:

The intra class correlation coefficient (ICC) across frequency for activity counts for all axes was 0.99 ($F_{53,1025}$ =41.3, p<0.001). The ICC across frequency for the X, Y and Z axes were 0.99 ($F_{17,341}$ =35.2, p<0.001), 0.99 ($F_{17,341}$ =38.8, p<0.001) and 0.99 ($F_{17,341}$ =38.6, p<0.001) respectively. ICC's did not differ when all 23 monitors were included (Table 5). Inter-instrument coefficients of variation (CV) showed greatest variability at 2.1 Hz, this reduced as frequency increased regardless of whether outliers were included or not (Table 6).

	N = 23	F Ratio	N = 19	F Ratio
X	0.99	$F_{17,413} = 28.2$	0.99	$F_{17,341} = 35.2$
Y	0.99	$F_{17,413} = 38.8$	0.99	$F_{17,341} = 38.8$
Z	0.99	$F_{17,413} = 33.0$	0.99	$F_{17,341} = 38.6$

Table 5. Intra-class correlation coefficients across frequency for each axis, including all 23 monitors and excluding the four outliers, all significant p < .001.

Table 6. Inter-instrument coefficient of variation (CV, %) for the mean activity counts at each axis and frequency, including all 23 monitors and excluding the four outliers.

2.1 Hz		5.1 Hz		10.2 Hz	
<i>N</i> = 23	N=19	<i>N</i> = 23	N = 19	<i>N</i> = 23	N=19
22.6	21.9	6.3	6.3	5.5	4.2
33.2	26.7	11.7	8.4	10.7	7.2
38.5	22.3	24.3	9.0	23.7	7.1
	2.1 I $N = 23$ 22.6 33.2 38.5	2.1 Hz $N = 23$ $N = 19$ 22.621.933.226.738.522.3	2.1 Hz5.1 I $N = 23$ $N = 19$ $N = 23$ 22.6 21.9 6.3 33.2 26.7 11.7 38.5 22.3 24.3	2.1 Hz5.1 Hz $N = 23$ $N = 19$ $N = 23$ $N = 19$ 22.621.96.36.333.226.711.78.438.522.324.39.0	2.1 Hz5.1 Hz10.2 Hz $N = 23$ $N = 19$ $N = 23$ $N = 19$ $N = 23$ 22.621.96.36.35.533.226.711.78.410.738.522.324.39.023.7

Intra-Instrument variability:

Intra-instrument CV showed greatest variability at 2.1 Hz along the Y axis compared to the X and Z axes regardless of whether all 23 monitors were included or not. The variability reduced as the frequency increased for all axes (Table 7).

	2.1 I	Iz	5.1 I	lz	10.2 Hz		
	N = 23	N=19	N = 23	N=19	<i>N</i> = 23	N = 19	
<u> </u>							
X	1.8 - 47.1	3.1 - 14.9	0.4 - 1.7	0.3 - 1.7	0.3 - 2.3	0.3 - 2.3	
	(10.6 ± 11.5)	(7.2 ± 2.1)	(0.0 ± 0.2)	(0,0,1,0,2)	(1.1 ± 0.6)	(1,1)	
	(10.0 ± 11.3)	(7.3 ± 3.1)	(0.9 ± 0.3)	(0.8 ± 0.3)	(1.1 ± 0.0)	(1.1 ± 0.0)	
\mathbf{v}	27-678	27-562	05 - 18	0.5 - 1.8	0.2 - 2.8	02 - 23	
	2.7 - 07.8	2.7 - 50.2	0.5 - 1.8	0.5 - 1.0	0.2 2.0	0.2 - 2.5	
	(15.4 ± 15.9)	(12.9 ± 12.1)	(1.1 ± 0.4)	(1.1 ± 0.4)	(1.2 ± 0.8)	(1.1 ± 0.8)	
			`				
Z	0-36.5	2.8 - 22.9	0.3 - 8.7	0.3 - 2.5	0.3 - 3.7	0.3 - 2.5	
	(12.9 ± 8.3)	(12.4 ± 6.4)	(1.7 ± 1.9)	(1.2 ± 0.6)	(1.2 ± 0.8)	(1.0 ± 0.6)	

Table 7. Intra-instrument coefficient of variation (CV, %) for each axis at each frequency, including all 23 monitors and excluding the four outliers, range (mean \pm SD).

Discussion

Without an accurate measure of physical activity, it is difficult to quantify relationships with health (Boreham and Riddoch, 2001). Additionally, when determining whether a population meets the national guidelines for physical activity it is necessary that the tool chosen to quantify physical activity is valid and reliable. The inter- and intra-monitor variability of the CSA uniaxial accelerometer has been determined and found to be acceptable (Metcalf et al., 2002). To our knowledge this is the first study to assess the technical variability of the RT3 accelerometer.

When assessing the variability of the output from the RT3s it is important that a range of scores obtained in human motion is used. Data from our laboratory (unpublished) have related RT3 counts to energy expenditure (METS) in children. In this study, a frequency of 2.1 Hz (acceleration = 0.057g) resulted in a mean activity count of 100.01 cts.min⁻¹ which reflected activities lower than 3 METs (e.g. crayoning), at 5.1 Hz (acceleration = 0.219g) a mean of 871.06 cts.min⁻¹ again reflected activities lower than 3 METs (e.g. playing catch), and at 10.2 Hz (acceleration = 0.414g) a mean of 1741.06 cts.min⁻¹ reflected activities of 3-6 METs, (e.g. walking). It must be noted that the higher frequencies used for testing are possibly out of the range of human motion, although the output amplitude is still within the range; however, frequencies of 5.1 to 10.2 Hz are in the dynamic capacity of the monitor and avoid the sampling issues discussed earlier in the method. Therefore, the results from the higher frequencies are relevant as it is the technical variability of the monitor being assessed. However, in terms of human motion the results at the 2.1 Hz frequency are perhaps most interesting.

At 2.1 Hz, the axes did not differ. At higher frequencies the Y axis consistently read higher than both the X and Z axes. On closer examination of the data this was the case for virtually all of the RT3s tested. Whether this is also the case for other RT3 monitors needs further investigation.

Intra-class correlation coefficients are impressive for the total sample and for individual axes (r = 0.99), reflecting the strong relationship between axes across all frequencies and across RT3s. The high inter- and intra-monitor coefficient of variation (CV) reported reflects the greater variability within and between RT3s at 2.1 Hz, compared to 5.1 and 10.2 Hz.

Although no instrumentation or test procedures are stated, Stayhealthy report data on their website (www.Stayhealthy.com) from the central 30 minutes of 40 minutes of testing using six randomly chosen monitors The activity counts achieved were on average 1794.26 (\pm SD, \pm 48.6), 1380.42 (\pm 34.6) and 1444.56 (\pm 16.7) cts.min⁻¹ at the X, Y and Z axes, respectively. These activity counts are comparable to the activity counts achieved at the 10.2 Hz frequency in the present study (1683.4 \pm

71.3, 1847.9 ± 132.2 and 1691.9 ± 119.7 cts.min⁻¹ for X, Y and Z axes, respectively). However, only inter-axes comparisons can be made and no other comparison is possible between the two studies as testing procedures were not made available.

Inter-instrument CV for the company data are impressive at less than 2.7 %, 2.5 % and 1.2 % for X, Y and Z respectively, in comparison to less than 5.3 % for both fast and medium speeds using the CSA uniaxial accelerometer (Metcalf et al., 2002) and less than 7.2 % at 10.2 Hz using the RT3 triaxial accelerometer. The relatively high CVs obtained in the current study may be inflated by the individual vibration of accelerometers in turn compared to the 'Stayhealthy' procedure of simultaneous shaking of all six RT3s. Details of the instrumentation, test procedures and frequency 'Stayhealthy' used when testing the RT3s are not available. It is acknowledged a larger sample of RT3 monitors would provide more conclusive results.

Inclusion of the four outlying monitors did not change any results. Intra-class correlation coefficients did not differ maintaining the strong relationship between axes across all frequencies and across all RT3s. However, in all cases the CV was reduced post exclusion (excluding inter-instrument CV on the X axis at 5.1 Hz, where there was no change).

This study did not test the robustness of the RT3 accelerometers. However, from a sample of 24 accelerometers, four failed (one due to water damage) during six months of use with children in physical activity research. In comparison Metcalf et al. (2002) reported that only one of the original 24 CSA uniaxial accelerometers failed during 12 months of use. As the monitors used in the present study were six months old the results presented cannot be generalized to newly bought RT3 activity monitors. It must also be acknowledged that the holster (provided by the company with each RT3 monitor for the attachment to a waist band), was attached firmly to the vibrator

plate and, even at the highest test frequency, can be regarded as a rigid structure (i.e. the holster was moving with the plate and not contributing some 'extra' vibration of its own). However, it cannot be ruled out that there was some movement of the RT3 within its holster during the high frequency measurement, although it was never observed to have moved at the end of a test.

In conclusion, the inter- and intra-instrument variability of the RT3 accelerometer has been assessed. The use of the activity monitor has research potential, allowing intensity, frequency and duration of physical activity to be measured. However, inter- and intra-monitor variability exists. Additionally, the Y axis reads consistently higher than the X and Z axes at 5.1 and 10.2 Hz. Therefore, it is recommended that all studies using the RT3 accelerometer perform trials to identify any outlying monitors and to assess the inter-monitor variability of RT3s. This level of quality control would ensure confidence in data obtained.

Future research should assess the technical reliability of the RT3s using apparatus with a larger field of movement, generating results for more direct comparison with human movement. Additionally, accelerometers should be tested repeatedly to allow comparisons on several occasions producing measures of validity and reliability. Ideally, this could lead to the publication of a standard procedure recommending how often the RT3 activity monitors need to be tested. Following controlled reliability tests the RT3s should be tested in a lab and field based environment on human subjects.

Chapter 5

Study 3

⁴Inter-monitor variability of the RT3 accelerometer during typical

physical activities.

⁴ This study is published in Medicine and Science in Sports and Exercise: Powell, S.M. and Rowlands, A.V. Inter-monitor variability of the RT3 accelerometer during typical physical activities. *Med. Sci Sports Exerc.*, 36:324-30, 2004.

Abstract

Purpose: The purpose of this study was to evaluate the reliability and variability of eight RT3 accelerometers. Methods: The RT3s (four on the right and four on the left hip) were subjected to two repeated trials of six activities: rest; walking (4 and 6 km.h ¹); running (8 and 10 km.h⁻¹); sit-stand position (20 min⁻¹). One person performed all trials (female: age 24 yrs, height 158.0 cm, mass 48.2 kg). Each activity lasted 12 minutes. The middle 10 minutes were taken from each 12 minute trial and used as the output measure (cts.min⁻¹). Data were analysed for activity (6), monitor (8) and trial (2) effects using four 3-way ANOVAs: vector magnitude; X (vertical); Y (anterioposterior); and Z (mediolateral) axes. Results: Inter-monitor coefficient of variation was <6% during locomotive activities. However, this increased to 8-25% during sit-stand activity. A 3-way interaction was found for vector magnitude (F_{35,315}=88945.7, p<.001), Y (F_{35,315}=978435.6, p<.001) and Z axes (F_{35,315}=103802.8, p < .001). A 2-way activity*monitor interaction was found for the X axis $(F_{35,315}=1037787.0, p < .001)$. Follow-up tests revealed no differences between trial one and two for vector magnitude, X and Z axes. One monitor recorded significantly lower activity counts in trial one compared to trial two along the Y axis. Inter-monitor differences were evident at 4, 6, 8 and 10 km.h⁻¹ for the Y and Z axes, and at 6, 8 and 10 km.h⁻¹ for the vector magnitude and X axis. Conclusions: Variability between monitors at each activity increased as intensity increased. Reliability of the RT3 is good. However, inter-monitor variability exists. The X axis of the RT3 accelerometer showed the least variability and was the most reliable. Therefore, the RT3 monitor may be most accurate as a uniaxial accelerometer. It is recommended that intermonitor variability and reliability of RT3s on each axis be assessed prior to use.

Introduction

Physical activity is recognized as a tool that can delay or prevent the onset of many chronic health conditions (Bouchard et al., 1994). Quantification of relationships between physical activity and health are often difficult due to indirect assessment e.g. historical recall, diaries or questionnaires (Rowlands et al., 2000). This has led to the need for objective measures that can be used to assess physical activity in the natural environment. Conceptually, assessment of physical activity using monitors that directly measure movement is attractive. Activity monitors however, are unable to detect the metabolic cost associated with standing, upper body movements, static work, vertical lift and changes in gradient (Bassett, 2000). Despite these limitations activity monitors have been shown to be a valid measure of physical activity (Eston et al., 1998; Trost et al., 1998).

Accelerometers measure the accelerations and decelerations of movement. The capture of intensity, frequency and duration information is enabled by a time sampling mechanism. The Computer Science and Applications, Inc. activity monitor (CSA model 7164, also known as the actigraph) is frequently used to assess physical activity in free living populations (Melanson et al., 2000; Tudor-Locke et al., 2002). It is a small, lightweight uniaxial accelerometer measuring movement along a vertical plane and is capable of storing activity data for up to 22 days. The CSA has been validated against oxygen consumption during typical children's activities (Eston et al., 1998) and been shown to be a technically reliable tool, with an inter-monitor coefficient of variation (CV) of less than 5% and an intra-monitor CV of less than 2% (Metcalf et al., 2002). Laboratory testing revealed no significant difference between two CSA accelerometers worn simultaneously at each of three treadmill speeds (3, 4 and 6

km.h⁻¹) and a high inter-monitor reliability coefficient when averaged over the three speeds (r = 0.87, Trost et al., 1998).

A three dimensional measure of activity (vertical, anterioposterior and mediolateral) is potentially important when assessing children's activities such as climbing and playing (Janz et al., 1995). Eston et al. (1998) showed that a triaxial accelerometer (TriTrac-R3D) was a more accurate predictor of oxygen uptake (scaled for body size) in children across a variety of activities than a uniaxial accelerometer (CSA). The TriTrac-R3D has been successfully validated against energy expenditure measured by indirect calorimetry in the laboratory (mean r=0.86 over a range of lab based activities, Welk et al., 2000) and in the field (r=0.62, Hendleman et al., 2000). A limitation associated with the TriTrac-R3D is its bulky nature ($120 \times 65 \times 22$ mm, 168g). The RT3 triaxial accelerometer is relatively small ($71 \times 56 \times 28$ mm, 65.2g), and replaced the TriTrac-R3D as a more researcher and user friendly device. The RT3 collects activity data in 1 or 60 second epochs and stores activity data for up to 21 days.

To our knowledge no studies concerning the validity of the RT3 accelerometer have been published. In our own laboratory, the RT3 has been successfully validated against oxygen uptake in both children and adults, over a range of regulated and nonregulated activities (r = 0.87, Rowlands et al., in press). When tested on a vibrating jig at 2.1 Hz (0.135g), 5.1 Hz (0.219g) and 10.2 Hz (0.414g) intra-monitor variability decreased as intensity increased (CV= 2-56%, 0-3% and 0-3%, respectively). Intermonitor variability showed the same pattern (CV= 22-27%, 6-9% and 4-7% respectively). Activity counts recorded were consistent between axes at 2.1 Hz. However; the Y axis recorded significantly higher activity counts at 5.1 and 10.2 Hz than the X and Z axes. Due to the large coefficients of variation for both intra- and inter-monitor variability at 2.1 Hz further testing has been recommended (Powell et al., 2003).

To date no study has assessed the inter-monitor variability of the RT3 accelerometer during standardised typical physical activities. Hence, the purpose of this study was to investigate the reliability and inter-monitor variability of the RT3, along each orthogonal axis of motion, over a range of activities.

Methods

Instrumentation.

The RT3 accelerometer.

The RT3 (Stayhealthy, Inc., Monvovia, CA, USA) is a small ($68 \times 48 \times 18$ mm), lightweight (65.2g), battery-powered monitor used as an experimental tool for measuring the physical activity of people. It is worn clipped to the waistband as an 'accessory' during waking hours. Depending on its mode of operation, it can record data for up to 21 days, which is then downloaded to a PC for display and statistical processing. The sensor in the RT3 is an accelerometer sensitive along three orthogonal axes, known as X (vertical), Y (anterioposterior) and Z (mediolateral). The acceleration is measured periodically, converted to a digital representation and processed to obtain an 'activity count' which is stored in memory.

The RT3 has four modes of operation: mode one samples and stores activity counts on individual axes at a 1 second (s) epoch; mode two samples and stores vector magnitude ($Vm = [X^2 + Y^2 + Z^2]^{0.5}$) activity counts at a 1 s epoch; mode three samples and stores accumulated activity counts on individual axes over a 60 s epoch; and mode four samples and stores accumulated vector magnitude activity counts over a 60 s epoch. The latter two modes store less detail about activity but are more economical in their use of memory, allowing longer duration experiments to be performed. Epoch

duration of 60 seconds is generally used in the field and all vectors were needed for this study, thus mode 3 was selected.

Eight RT3 accelerometers were selected randomly from a sample of 20, which were all 6 months old and previously used in the field. The RT3s were initialised via a computer interface, simultaneously started and split into two sets of four. The RT3s were placed back-to-front, all facing forward and in sets of four securely taped together. This gave two blocks of monitors which were attached to a belt, positioning each block above either the left or right hip at waist level.

Procedures

The reliability and inter-monitor variability of the RT3s was assessed during two trials of six activities: rest; walking (4 and 6 km.h⁻¹); running (8 and 10 km.h⁻¹, performed on an electronically driven treadmill, Powerjog, model JM200, Sport Engineering Ltd., UK); and a repeated sit to stand (metronome controlled at 40 beats per minute). Each activity was performed for 12 minutes, with at least a 10 minute break in-between each activity. The two trials were performed two days apart. After each trial the RT3 monitors were removed and data downloaded. The first and last minute of each 12 minute bout was deleted, leaving 10 minutes at each activity for each trial. Data output were counts per minute (cts.min⁻¹). All monitors tested were worn at the same time by one participant (female: age 24 yrs, height 158.0 cm, weight 48.2 kg) in a laboratory setting.

Pilot testing using two RT3 accelerometers, known to have high inter-monitor consistency (technical variability measured previously in an electronically controlled environment, Powell et al., 2003) tested the left to right hip placement effects. Over one trial of six minutes, at all previously mentioned activities, there were no significant differences between left and right hip placements ($F_{1.697}=1.5$, p=0.3). This

supports previous research which also resulted in no left to right hip differences in the laboratory (Trost et al., 1998). Therefore, it was assumed inter-monitor variability could be assessed without reference to hip placement.

Statistical Analysis

Descriptive statistics were calculated for each activity at trial one and trial two. Inter-monitor coefficient of variation (CV) for each activity at each trial was calculated. A 3-way mixed model ANOVA (activity*monitor*trial) was performed to examine the inter-monitor variability and reliability of the RT3s for each vector. Significant 3-way interactions were followed-up using two, 2-way mixed model ANOVAs (activity*monitor): one for trial one and one for trial two. Significant 2-way interactions were followed-up using the adapted Tukeys test for repeated measures (Stevens, 1996). Where necessary, degrees of freedom were adjusted according to the Greenhouse Geisser correction due to violation of the assumption of sphericity. Between activities, monitor differences are reported as percentages of possible monitor pairings (64 total possible pairings) that did not differentiate between two consecutive activities (Table 8). Within activity and within trial, monitor differences are reported as percentages of possible monitor pairings (28 total possible pairings) that were significantly different within each activity or trial (Table 9, Figure 11).

All statistical procedures were carried out for vector magnitude, X, Y and Z axes. Due to the number of analyses an alpha level of .01 was used for all statistical procedures to control for an increased risk of type I error.

Results

Descriptive data are shown in Table 10. Inter-monitor CV (SD/mean * 100, Table 11) showed low variation (<6%) for all axes during locomotor activities (4-10 km.h⁻¹) however, relatively high variation was evident during sit-stand (8-25%). Due to the very low mean score at rest the CV was inflated and not considered meaningful, therefore, it is not presented in Table 11.

Table 8. Percentage of possible pairings of monitors that did not differentiate between activities (rest and sit-stand, sit-stand and 4 km.h⁻¹, 4 and 6 km.h⁻¹, 6 and 8 km.h⁻¹ and 8 and 10 km.h⁻¹).

	Trial	Rest-SS	SS-4 km.h ⁻¹	4-6 km.h ⁻¹	6-8 km.h ⁻¹	8-10 km.h ⁻¹
Vm	1	100	0	0	0	16
	2	100	0	0	0	34
X	1	100	0	0	0	31
	2	100	0	0	0	36
Y	1	100	2	19	16	44
	2	100	2	14	20	38
Z	1	100	11	16	17	61
	2	100	11	16	17	56

Vm = vector magnitude,

SS = sit-stand.

	Trial	Rest	4 km.h ⁻¹	6 km.h ⁻¹	8 km.h ⁻¹	10 km.h ⁻¹	Sit-Stand
Vm	1	0	0	25	75	82	0
	2	0	0	21	64	79	0
X	1	0	0	4	46	79	0
	2	0	0	11	43	75	0
Y	1	0	11	50	89	93	0
	2	0	32	43	82	96	0
Z	1	0	25	54	89	86	0
	2	0	39	57	93	93	0

Table 9. Percentage of possible pairings of monitors significantly different within each activity.

Vm = vector magnitude.

	Trial	Rest*	4 km.h ⁻¹	6 km.h ⁻¹	8 km.h ⁻¹	10 km.h ⁻¹	Sit-stand
Vm	1	0.3	1158.2	2404.5	4807.9	5590.2	158.1
		± 0.3	± 36.8	± 50.6	± 182.5	± 86.0	± 22.8
	2	0.3	1124.5	2472.3	4855.1	5597.4	192.8
		± 0.3	± 32.9	± 49.7	± 112.6	± 121.4	± 16.8
х	1	0.0	745.5	1562.5	2838.4	3484.9	68.3
		± 0.0	± 29.7	± 29.7	± 104.4	± 56.6	± 10.1
	2	0.1	780.4	1585.7	2825.5	3397.1	72.2
		± 0.3	± 24.0	± 38.3	± 89.8	± 61.7	± 9.5
Y	1	0.0	491.7	945.3	1913.1	2483.7	62.8
		± 0.0	± 24.4	± 56.9	± 41.1	± 98.0	± 16.1
	2	0.0	542.7	1005.5	1889.9	2463.7	88.9
		± 0.0	± 17.7	± 31.6	± 83.8	± 97.7	± 12.3
Z	1	0.2	707.2	1555.0	3166.4	3202.9	122.9
		± 0.3	± 24.4	± 77.2	± 141.7	± 64.2	± 15.5
	2	0.3	785.6	1613.7	3215.5	3366.5	149.9
		± 0.3	± 27.2	± 72.7	± 88.5	± 131.6	± 13.3

Table 10. Activity by vector descriptive statistics (cts.min⁻¹, mean \pm SD).

Vm = vector magnitude,

*rest not significantly different from sit-stand, p>.01.

	Trial	4 km.h ⁻¹	6 km.h ⁻¹	8 km.h ⁻¹	10 km.h ⁻¹	Sit-Stand
Vm	1	3.2	2.1	3.8	1.5	14.4
	2	2.9	2.0	2.3	2.2	8.7
Х	1	4.0	1.9	3.7	1.6	14.8
	2	3.2	2.4	3.2	1.8	13.2
Y	1	5.0	6.0	2.2	3.9	25.6
	2	3.3	3.1	4.4	4.0	13.8
Z	1	3.5	5.0	4.5	2.0	12.6
	2	3.5	4.5	2.8	3.9	8.9

Table 11. Inter-monitor coefficients of variation by activity (CV, %).

Vm = vector magnitude.

Vector magnitude. A 3-way interaction (activity*monitor*trial) was found ($F_{35,315}$ =88945.7, p<.001). Follow-up 2-way ANOVAs revealed significant activity*monitor interactions, at both trial one ($F_{35,315}$ =131.6, p<.001) and trial two ($F_{35,315}$ =96.7, p<.001, Figure 11). At both trial one and trial two, all activities were significantly different from each other with the exception of rest and sit-stand, and a percentage of monitors not differentiating between 8 and 10 km.h⁻¹ (16% at trial one and 34% at trial two, Figure 11, Table 8). Within activities, there were no significant differences between monitors at rest, 4 km.h⁻¹ or sit-stand, at trial one or trial two. However, as intensity increased (6 to 10 km.h⁻¹) the inter-monitor difference increased (21 to 82%, Figure 14, Table 9).

There was no activity*trial interaction however, there was a monitor*trial interaction ($F_{7,63}=3.0, p<.01$). Follow-up tests revealed 21% (trial one) and 18% (trial

two) of possible monitor pairings were significantly different within trial (Figure 15). No monitors were significantly different between trial one and two.



Figure 14. Monitor by activity: vector magnitude (mean \pm SEM). Activities 4, 6, 8 and 10 = km.h⁻¹, SS = sit-stand.

* = no significant difference between RT3 monitors at rest, 4 km.h^{-1} or sit-stand.





X axis. There was no 3-way interaction and no activity*trial or monitor*trial interactions. There was however, a significant activity*monitor interaction $(F_{35,315}=1037787.0, p<.001, Figure 16)$. All activities were significantly different from each other with the exceptions of rest and sit-stand, and 23% of the possible monitor pairings not differentiating between 8 and 10 km.h⁻¹ (Figure 16, Table 8). Within activities, there were no significant differences between monitors at rest, 4 km.h⁻¹ or sit-stand. However, as intensity increased (6 to 10 km.h⁻¹) the inter-monitor difference increased (4 to 75%, Figure 16, Table 9). No difference across trials was shown for the X axis. However, for consistency of presentation of results tables 8 and 11 present data for each trial separately.



Figure 16. Monitor by activity estimated statistics: X axis (mean \pm std. error). Activities 4, 6, 8 and $10 = \text{km.h}^{-1}$.

SS = sit-stand.

* = no significant differences between RT3 monitors at rest, 4 km.h⁻¹ and sit-stand.

Y axis. A 3-way interaction was found ($F_{35,315}=978435.6$, p<.001). Follow-up 2-way ANOVAs revealed significant activity*monitor interactions, at both trial one ($F_{35,315}=278.4$, p<.001) and trial two ($F_{35,315}=183.8$, p<.001, Figure 17). At both trial

one and two, rest and sit-stand were not significantly different from each other, and a proportion of the possible monitor pairings, increasing with intensity, did not differentiate between sit-stand and 4 km.h⁻¹, 4 and 6 km.h⁻¹, 6 and 8 km.h⁻¹ and 8 and 10 km.h⁻¹ (2 to 44%, Figure 17, Table 8). Within activities, there were no significant differences between monitors at rest or sit-stand, for trial one or trial two. However, as intensity increased (4 to 10 km.h⁻¹) the inter-monitor variability increased (11 to 96%, Figure 17, Table 9).



Figure 17. Monitor by activity: Y axis (mean \pm std. error).

Activities 4, 6, 8 and $10 = \text{km.h}^{-1}$,

SS = sit-stand.

* = no significant differences between RT3 monitors at rest and sit-stand.

There was no activity*trial interaction however, there was a monitor*trial interaction ($F_{7,63}=52.9$, p<.001). Follow-up tests revealed 14% (trial one) and 25% (trial two) of possible monitor pairings were significantly different within trial (Figure 18). There was also one monitor which revealed significantly lower activity counts at trial one compared to trial two.

B



Figure 18. Monitor by trial: Y axis (mean \pm std. error).

* = significant difference across trials for RT3 monitor 2.

Z axis. A 3-way interaction was found ($F_{35,315}=103802.8$, p<.001). Follow-up 2-way ANOVAs revealed significant activity*monitor interactions, at both trial one ($F_{35,315}=189.9$, p<.001) and trial two ($F_{35,315}=229.3$, p<.001, Figure 19). At both trial one and two, rest and sit-stand were not significantly different from each other, and a proportion of the possible monitor pairings, increasing with intensity, did not differentiate between sit-stand and 4 km.h⁻¹, 4 and 6 km.h⁻¹, 6 and 8 km.h⁻¹ and 8 and 10 km.h⁻¹ (11 to 61%, Figure 19, Table 8). Within activities, there were no significant differences between monitors at rest or sit-stand, for trial one or trial two. However, as intensity increased (4 to 10 km.h⁻¹) the inter-monitor variability increased (25 to 93%, Figure 19, Table 9). There was no activity*trial or monitor*trial interaction.


Figure 19. Activity by monitor: Z axis (mean \pm std. error).

Activities 4, 6, 8 and $10 = \text{km.h}^{-1}$,

SS = sit-stand.

* = no significant differences between RT3 monitors at rest and sit-stand.

Discussion

To our knowledge this is the first study to evaluate the reliability and variability of the RT3 accelerometer, across a variety of laboratory based physical activities. Without an accurate measure of physical activity, it is difficult to quantify relationships with health (Boreham and Riddoch, 2001). This study shows individual RT3 monitors were reliable over trials, with the exception of one monitor on the Y axis. The RT3s largely differentiated between low activity intensities (e.g. sit-stand, 4-6 km.h⁻¹), however differentiation decreased as activity intensity increased (e.g. 8-10 km.h⁻¹). Considerable inter-monitor differences within activities were apparent on all axes. The X axis of motion revealed the least variability between monitors. Due to high variability on the Y and Z axes, the vector magnitude variability was also high.

Reliability is an important issue when using activity monitors to record physical activity over a period of time. The vector magnitude (mean count at trial one compared to (cf.) trial two, 2353 cf. 2394 cts.min⁻¹), X (1450 cf. 1444 cts.min⁻¹) and Z (1459 cf. 1522 cts.min⁻¹) axes of motion were shown to be reliable over trials. The Y axis (983 cf. 999 cts.min⁻¹) also showed reliability over trials, with the exception of one monitor which elicited significantly higher activity counts at trial one cf. trial two. However, inter-monitor differences within each trial highlight the importance of matching the same RT3 to a subject performing repeated trials.

When considering the variability of activity monitors, a range of activity intensities should be considered to adequately test the assumption that the monitor can differentiate between important cut-off points of physical activity. Unpublished data from our laboratory have related RT3 counts to energy expenditure (METS) in children (Table 12). Rest (vector magnitude \pm SD, 0 \pm 0 cts.min⁻¹) and sit-stand (176 \pm 16 cts.min⁻¹) reflected less than 3 MET counts, compared to 4 km.h⁻¹ (1202 \pm 31 cts.min⁻¹= moderate activity 3-6 METs), 6 km.h⁻¹ (2438 \pm 37 cts.min⁻¹= vigorous activity 6-9 METs), 8 km.h⁻¹ (4832 \pm 103 cts.min⁻¹= very hard activity >12 METs) and 10 km.h⁻¹ (5594 \pm 70 cnts.min⁻¹= very hard activity >12 METs). It is important to relate physical activity to known cut-off points for the relationship with health recommendations to be identified. Within each 10 minute trial, at each intensity, individual monitors were consistent in recording counts that did not vary outside one intensity classification.

Activity intensity	Activity counts	MET value	
Low	0 - 950	0 - 2.9	
Moderate	951 - 2300	3 - 5.9	
Vigorous	2301 - 3200	6 - 8.9	
Hard	3201 - 4100	9 - 11.9	
Very hard	>4101	>12	

 Table 12. The relation of activity intensity to accelerometer counts and MET
 equivalents.

Activity counts and MET equivalents taken from Rowlands et al. (2004).

Rest and sit-stand were not found to be significantly different by any of the axes of motion, due to the very low activity counts elicited by each activity. The vector magnitude and X axis differentiated between all other activities well, excluding an overlap between monitors at 8 and 10 km.h⁻¹. However, the Y and Z axes revealed overlap between monitors at all activities. As intensity increased the percentage of possible pairings which did not differentiate between activities increased. When assessing inter-monitor variability of two CSA uniaxial accelerometers at three speeds (3, 4 and 6 km.h⁻¹) Trost et al. (1998) found the CSA differentiated between the three speeds tested. However, high intensities of activity (above 6 km.h⁻¹) and very low intensities were not examined and therefore cannot be compared. These high and very low intensities of activity were where the RT3 showed least differentiation.

Accerometers have previously been reported to have difficulty in distinguishing between running speeds above 8 km.h⁻¹ (Brage et al., 2003). Brage et al. (2003) reported the results of 12 male subjects who performed three treadmill trials containing 5 minute blocks at two walking speeds (3 and 6 km.h⁻¹) and five running speeds (8, 9, 10, 12 and 14 km.h⁻¹) whilst wearing four CSA uniaxial accelerometer,

two above each hip. In support of the present study, activity counts on the vertical axis rose linearly with speed up until 8 km.h⁻¹, however further increments in running speed did not increase the accelerometer output. It has been suggested that the observed levelling-off in the CSA output during running may be largely due to biomechanical characteristics i.e. that average vertical acceleration is relatively constant across running speed (5, 8, 11). Bouten et al. (1994) reported VO₂ was better predicted by the anterioposterior axis of the tri-axial tracmor despite the major acceleration component occurring along the vertical axis. They recommended the use of the Y axis to differentiate between running intensities. However, this is not supported by the results of the present study as none of the vectors differentiated between 8 and 10 km.h⁻¹.

When using a number of activity monitors in a single study it is important the inter-monitor variability is minimal, if not comparison of activity levels between participants is not possible. Within each of the given activities the X axis was shown to have the least inter-monitor variability, compared to the Z axis which elicited the most variability at both 6 and 8 km.h⁻¹, and the Y axis at 10 km.h⁻¹. In all cases, as the activity became more intense the variability between the RT3s increased. Only at rest and sit-stand was there consistently no variability between the RT3s due to the low activity counts elicited. Trost et al. (1998) found no differences between two CSA monitors at any of the three speeds tested (3, 4 and 6 km.h⁻¹). However, only two monitors were tested and only at relatively low speeds. The present study highlights the higher variability elicited between monitors at high speeds. Coefficient of variation (CV) during locomotor activities was low (<9%) and similar to that reported for the CSA (<5%, Metcalf et al., 2002) and the RT3 (<9%, Powell et al., 2003) during controlled lab based studies. Despite this, quite large inter-monitor differences were apparent at high intensities. The increase in variability with speed is also supported by

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Brage et al. (2003), inter-monitor differences in relation to the common mean were within \pm 30%, however as speed increased so did CSA inter-monitor differences. Therefore, when using more than one RT3 accelerometer in a single study, intermonitor variability must be assessed.

Results of this study support research performed in our laboratory concerning the technical variability of the RT3 (Powell et al., 2003). Twenty-three RT3 accelerometers were subjected to a specific vibration along each sensitive axis in isolation, using a motorized vibration table which produced frequencies of 2.1 Hz (0.057g), 5.1 Hz (0.219g) and 10.2 Hz (0.414g), respectively. Results revealed no differences in activity counts on the X, Y and Z axes at 2.1 Hz however, the activity counts recorded along the Y axis were significantly higher than those at the X and Z axes at 5.1 and 10.2 Hz. Intra- and inter-monitor coefficients of variation decreased as frequency increased, supporting the decrease in CV with the increase in frequency of movement (e.g. sit-stand to running) in the present study. In both studies the X axis performed well in comparison to other axes of motion.

Although it has been suggested that activity monitors should be placed as close to the centre of gravity as possible (Westerterp, 1999), placement at the waist over one hip has proved to be popular, possibly for comfort reasons (Nilsson et al., 2002). The effect of the placement of the RT3s (left versus right hip) tested in a pilot study prior to data collection, found no differences in activity counts recorded during all aforementioned activities between the left and right hip placement. This supports Trost et al. (1998) who found no difference between left and right hip placement and high correlations between both left and right hip placement with energy expenditure during treadmill exercise. This is in contrast to Fairweather et al. (1999), who found significant differences (3%) between left and right hip placements with the CSA uniaxial accelerometer. However, the magnitude of the difference between left and

right hips placement was similar to the inherent imprecision of inter-monitor difference found under in-vitro conditions. Although placement effects were considered prior to data collection they cannot be ruled out in the present study. However, visual examination of the data indicated no placement bias. In future research of this nature, it may be advisable to control for placement effects prior to and during data collection.

The possibility of differences in human motion from trial one to trial two cannot be disregarded. However, all axes of motion were found to be reliable over trials, with the exception of one monitor on the Y axis. Therefore, it can be assumed the magnitudes of any differences inferred by human motion are smaller than those from each RT3 accelerometer. The possibility of individual monitor movement is also acknowledged. As each set of four RT3 accelerometers were taped firmly together and tightly to the waist, it is likely any movement would have been minimal. It is also possible a 60 second epoch was too high for this study where each activity was performed for 12 minutes only. Nilsson et al. (2002) suggested shorter bursts of vigorous activity may be blunted when counts are summed over one minute. As each activity was performed at a constant rate for 12 minutes and the first and last minute discarded, this should not have been a problem. A practical limitation was that only eight RT3s were tested, all were six months old and previously used in the field. It would be of interest to use a larger sample of new RT3s to see whether the results are comparable. This may reveal aspects of long term reliability and whether intermonitor variability increases with monitor age.

In conclusion, with sedentary groups the RT3 accelerometer may be an adequate monitor for obtaining an informative picture of activity. However, ideally and especially when measuring active populations, every laboratory should satisfy itself that the inter-monitor variability of their RT3s is acceptable before use.

Acceptable parameters of variability depending on the nature of the research to be carried out should be outlined by laboratories using RT3 monitors. The X axis of motion was reliable over time, differentiated well between activities and elicited the least variability within each activity. The RT3 accelerometer is a user friendly tool to monitor physical activity and may be considered a reliable tool at least as a uniaxial monitor, recording data along a vertical axis of motion only. Future research should use a larger sample of RT3s not previously used in the field. They should be tested over a long period of time with a variety of activities both in the laboratory and in the field. It would be beneficial to replicate this study with a child participant using the same protocol to assess both uni-axial and tri-axial accelerometers.

Chapter 6

Study 4

⁵Accelerometer sampling effects on recorded physical activity in

children

⁵ This study has been submitted to Pediatric Exercise Science.

Abstract

Purpose: To evaluate the influence of accelerometer time sampling interval on recorded measures of physical activity in children. Methods: Twenty-five children wore two RT3 accelerometers for six school hours. Activity was recorded at 60 s and 1 s epochs. Minutes in moderate, vigorous, hard and very hard activities were recorded. Results: The 60s epoch overestimated time in moderate and vigorous activity, but underestimated time in very hard and ≥vigorous activity. Conclusion: A 1s epoch should be used when assessing ≥vigorous activity. This is important when assessing the relationship between bone health and activity as bone mass is increased by short periods of intense activity.

Introduction

Without an accurate measure of physical activity, it is difficult to quantify relationships with health (Boreham and Riddoch, 2001). In addition, it is not possible to assess whether a population meets the national guidelines for physical activity unless the tool chosen to quantify physical activity is valid.

Accelerometers measure the accelerations of movement. A time sampling mechanism allows the capture of intensity, frequency and duration information. The three dimensions of assessment offered by triaxial accelerometry (X = vertical, Y = anterioposterior and Z = mediolateral) are potentially important when assessing children's physical activity, due to the greater variety of movements undertaken by children relative to adults. Eston et al. (1998) showed that a triaxial accelerometer (TriTrac-R3D) was a more accurate predictor of oxygen uptake in children, across a variety of activities, than a uniaxial accelerometer. The TriTrac-R3D has been successfully validated against energy expenditure measured by indirect calorimetry in the laboratory (r = .86, Eston et al., 1998). However, a limitation associated with the TriTrac-R3D is its bulky nature ($120 \times 65 \times 22$ mm, 168g). The RT3 accelerometer is much smaller ($71 \times 56 \times 28$ mm, 65.2g) than the TriTrac-R3D, and was introduced as a more researcher- and user-friendly device. The RT3 has been successfully validated against the criterion of oxygen uptake in both children and adults in our own laboratory (Rowland et al., 2004).

Accelerometers are often set at a sampling interval (epoch) of 60 seconds (Ainsworth et al., 2000; Cooper et al., 2000; Levin et al., 1999), which summarises all registered counts during this period maximising the memory capacity. However, children's activity patterns are spontaneous and intermittent in their nature (Rowlands et al., 1997). Bailey et al. (1995) reported that children engaged in very short bursts of intense physical activity interspersed with varying intervals of low and moderate intensity. The median duration of high intensity activities was found to be only 3 seconds with 95% lasting less than 15 seconds. Nilsson et al. (2002) showed the use of longer epochs may not capture high intensity activity accurately. Short bursts of very hard activity coupled with light activity may not accumulate enough counts to pass the threshold for the very hard intensity bracket and be resigned to a lower intensity bracket. The number of minutes recorded in high intensity activities decreased as the epoch setting increased from 5 seconds to 60 seconds. At epoch settings of 5, 10 and 20 seconds 11.7, 7.9 and 3.8 minutes, respectively, were recorded in very high intensity activities (\geq 9 METs) compared to only 1.3 minutes at the 60 second epoch. Therefore choice of epoch appears to be very important when assessing activity intensity. The RT3 has epoch settings of 1 s or 60 s only and recording time is very limited with the 1 s epoch (up to 9 hours, compared to up to 21 days with the 60 s epoch), therefore it is important to assess the extent of any misclassification of activity intensity when selecting the 60s epoch.

Short periods of intense activity are particularly important for bone health. High intensities of strain to the musculoskeletal system appear to be more important than the volume of activity to bone development (Parker, 1998). Therefore, underestimation of time spent in high intensity activities might mask relationships between physical activity and bone health.

The purpose of this study was to investigate the effect of epoch selection (60 second and 1 second) of the RT3 accelerometer on recorded habitual physical activity (total and minutes spent at varying intensities) in children.

Methods

Participants

Twenty-five children were recruited from a local primary school in North Wales. Of these participants, 15 were boys (mean \pm SD, age = 8.8 ± 1.1 yrs, mass = 32.9 ± 8.0 kg, height = 133.6 ± 6.9 cm) and 10 were girls (age = 8.5 ± 0.1 yrs, mass = 30.6 ± 4.3 kg, height = 132.4 ± 8.1 cm). Written informed consent was obtained from all parents and verbal consent from all children. Each participant was visited at school after assembly (9.30 am), where all procedures were explained. Height was measured to the nearest 0.1 cm using a free standing Seca stadiometer and body mass was measured to the nearest 0.1 kg using Seca scales.

Instrumentation

The RT3 (Stayhealthy, Inc., Monvovia, CA, USA) is a small (71 × 56 × 28 mm), lightweight (65.2g), battery-powered instrument used as an experimental tool for measuring physical activity. It is worn clipped to the waistband as an 'accessory' during waking hours. Depending on its mode of operation, it can record data for up to 21 days, which is then downloaded to a PC for display and statistical processing. The sensor in the RT3 is an accelerometer which measures acceleration periodically; it is then converted to a digital representation and processed to obtain an 'activity count' which is stored in memory. The exact relationship of the activity count to the acceleration (measured in m.s⁻² or g, where 1g = 9.81m.s⁻²) is not clear.

The RT3 has four modes of operation: mode one samples and stores activity counts on three individual orthogonal axes at 1 s epochs for up to three hours; mode two samples and stores vector magnitude (a measure combining all three axes of motion) activity counts at 1s epochs for up to nine hours; mode three samples and stores accumulated activity counts on individual axes over 60 s epochs for up to seven days; and mode four samples and stores accumulated vector magnitude activity counts over 60 s epochs for up to 21 days. The latter two modes store less detail about activity but are more economical in their use of memory, allowing longer duration experiments to be performed. Epoch duration of one minute is generally used in the field. The vector magnitudes (a culmination of the three vectors) at 60 s and 1 s epochs were used.

Physical activity assessment

From a possible nine RT3 accelerometers, each child wore two randomly selected accelerometers for up to six school hours. The accelerometers were strapped to a belt worn by the child. The accelerometers were taped together and positioned over the right hip: one monitor was programmed to record at a 60 s epoch and the other at a 1 s epoch. The accelerometers were initialised and downloaded via a computer interface and had no external controls that could be manipulated. Output measures for each monitor were total activity counts, minutes spent in very hard, hard, vigorous, moderate and low intensity activities. The definition and cut-off point used for each activity intensity are shown in Table 13.

Activity intensity	Activity counts	Activity counts	MET value	
	(60s epoch)	(1s epoch)		
Low	0 - 970	0 - 16	0-2.9	
Moderate	970 - 2333	17 – 39	3 - 5.9	
Vigorous	2333 - 3200	40 – 54	6 - 8.9	
Hard	3201 - 4100	55 – 69	9 - 11.9	
Very hard	4101+	70+	12+	

Table 13. The relation of activity intensity to accelerometer counts and MET equivalents.

Activity counts and MET equivalents taken from Rowlands et al. (2004).

Statistical Analysis

Descriptive statistics were calculated for all variables. Independent *t*-tests were used to examine gender differences. A series of six one-way repeated measures ANOVAs were performed to examine differences between monitors (60 s epoch and 1 s epoch) in time spent at the various activity intensities (light, moderate, vigorous, hard, very hard, above moderate). Where necessary, degrees of freedom were adjusted using the Greenhouse Geisser correction due to violation of the assumption of sphericity. Pearson's correlation coefficient was used to examine the relationship between total physical activity (1 s epoch) and bias between time recorded at the various intensities of activity by both the monitor set at a 1 s epoch and the monitor set at a 60 s epoch. Alpha was set at .05. This was adjusted to .01 for the series of oneway ANOVAs to account for the increased risk of type 1 error due to multiple tests.

To assess the extent to which total physical activity and the time recorded at each intensity agreed between the 60 s epoch and the 1 s epoch, levels of agreement were calculated as described by Bland and Altman (1986). The consensus of opinion

suggests that this is the most appropriate technique for the assessment of measurement agreement (Nevill and Atkinson, 1997). The average bias and 95% confidence interval for the bias were calculated for total physical activity, time recorded in light, moderate, vigorous, hard, very hard and above moderate activity. Systematic bias was shown by a correlation between bias and the average of the monitors for all measures except total physical activity and time recorded in very hard intensity activity. This systematic bias affects the accuracy of the limits of agreement. Log transformation of the data, as recommended by Bland and Altman (1986), did not solve this problem. Therefore the calculated limits of agreement for these measures will have a tendency to be too far apart (1986).

Results

Variable	Boys $(n = 15)$	Girls $(n = 10)$	Total $(n = 25)$
Age (years)	8.8 ± 1.1	8.5 ± 0.9	8.7 ± 1.0
Height (cm)	133.6 ± 6.9	132.4 ± 8.1	133.1 ± 7.3
Weight (kg)	32.9 ± 8.0	30.6 ± 4.3	32.0 ± 6.8
60 s epoch	228044 ± 49964	183809 ± 60908	210350 ± 57763
1 s epoch	246495 ± 50496	219421 ± 61484	235666 ± 55572

Table 14. Descriptive statistics (mean \pm SD).

There were no significant differences between the boys and girls on any of the descriptive variables (Table 14). There were no significant differences between monitors for minutes in light intensity activity ($F_{1,24} = 2.7, p = 0.11$), minutes in hard intensity activity ($F_{1,24} = 0.9, p = 0.35$) or minutes in \geq moderate intensity activity ($F_{1,24} = 2.5, p = 0.13$). However, the monitor with a 1 s epoch recorded significantly

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fewer minutes in moderate activity ($F_{1,24} = 11.6$, p < 0.01) and vigorous activity ($F_{1,24} = 20.7$, p < 0.01), and more minutes in very hard activity ($F_{1,24} = 63.5$, p < 0.01) and \geq vigorous activity ($F_{1,24} = 7.9$, p < 0.01), relative to the monitors with a 60 s epoch, (Figure 20).



Figure 20: Number of minutes recorded at each activity intensity by the 1s and 60 s epoch.

*Number of minutes recorded by the 1 s epoch monitor significantly different from number of minutes recorded by 60 s epoch monitor.

Total physical activity (1 s epoch) correlated significantly (r = -.0.71, p < 0.001) with the difference in time recorded at a very hard intensity by the 1 s and 60 s epoch monitors (60 s – 1 s epoch). The negative correlation indicates that the degree

of underestimation by the 60 s epoch increased as total physical activity increased. No other significant correlations between total physical activity and bias were evident.

The mean bias and 95% limits of agreement are shown in Table 15.

		Mean of the	Bias	95% limits of
		two monitors		agreement
60 s – 1 s epoch*	Total physical activity	223008	-25315	92614
	Light**	279.9	-5.3	31.6
	Moderate**	35.3	9.4	27.0
	Vigorous**	10.0	3.3	7.1
	Hard**	6.5	1.0	10.2
	Very hard	8.0	-8.6	10.6
	\geq moderate**	59.9	5.0	31.4

Table 15. Limits of agreement

*A negative bias indicates an underestimation by the 60 s epoch monitor relative to the 1 s epoch monitor. Conversely a positive bias indicates an overestimation by the 60 s epoch monitor.

** Bias significantly correlated with mean of the two monitors (p < 0.05), therefore limits of agreement may be inflated.

Mean bias was low for total and light intensity activity, though, increased with intensity. Individual differences were substantial, reflected by the large limits of agreement. The estimates of limits of agreement may be on the liberal side for all measures except total physical activity and very hard physical activity due to the systematic bias in difference between monitors.

Discussion

When assessing physical activity in the field using accelerometry, a 60 s epoch is generally selected due to memory limitations of shorter epochs. However, the accuracy of assessment of high intensity activity when using a 60 s epoch has been questioned (Nilsson et al., 2002). This study aimed to determine whether output measures of physical activity were affected by the duration of the epoch of the RT3.

Epoch effects

No significant differences were found in the number of minutes recorded during light (<3 METs) or moderate (3-5.9 METs) activities, when using the 60 s epoch and the 1 s epoch setting, supporting work with the CSA uniaxial accelerometer. This is supported by Nilsson et al. (2002) who found no significant difference between the number of minutes recorded at moderate intensity activity (<6 METs) by a 5 s epoch compared to a 60 s epoch. This suggests that activities which expend no more than 6 METs are usually maintained for over 60 seconds. Therefore, a 60 s epoch setting is adequate to capture these periods of movement.

Vigorous intensity activity (6-8.9 METs) was overestimated and very hard intensity activity (> 12 METs) underestimated by the 60 s epoch relative to the 1 s epoch setting. However, no significant difference was recorded between the epoch settings during hard (9-11.9 METs) activities. In contrast, Nilsson et al. (2002) reported an underestimation in the time recorded at high (6-9 METs, equivalent to our vigorous intensity) and very high (>9 METs, equivalent to our hard and very hard intensity activity combined) by a 60 s epoch setting relative to a 5 s epoch setting. No differentiation was made with activities exceeding 12 METs. However, when the number of minutes were combined in the present study to form the corresponding >9

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METs category, the 60 s epoch setting similarly underestimated time relative to the 1 s epoch (p < 0.001).

Children show sporadic activity patterns engaging in very short bursts of intense activity, typically lasting less than 15 seconds (Bailey et al., 1995); a 60 s epoch is unlikely to capture these short episodes. To be classified as very hard intensity activity when using an epoch setting of 60 s a count of >4101 must be accumulated during the 60 s. This may be due to prolonged very hard intense activity (> 12 METS) lasting for over 60s or very intense bouts of activity (> 12 METS) coupled with lower intensity motion. If the very hard intensity activities are not maintained or the bouts are not intense enough to accumulate >4101 counts, they will be resigned to a lower category activity intensity. If the child participates only in sporadic hard or very hard intensity activity it is likely to go undetected (Nilsson et al., 2002). This may also explain the overestimation during vigorous intensity activity. Activity classified as very hard by the 1 s epoch monitor may have been too short to be recorded as very hard or even hard by the 60 s epoch monitor and hence may be classified as vigorous. This would lead to the overestimation of vigorous activity and the underestimation of very hard activity.

Despite the lack of significant differences between time recorded at most intensities, levels of agreement analysis revealed wide individual variation for all but light intensity activity. For example: if the 1 s epoch monitor recorded 300 minutes light activity, the 95% limits for the 60 s epoch monitor indicate it could read between 263 and 326 minutes; 30 minutes moderate activity could read between 12 and 66 minutes; 10 minutes vigorous activity could read between 6 and 20 minutes; 10 minutes hard activity could read between 1 and 21 minutes; 10 minutes very hard activity could read between 0 and 12.6 minutes; 60 minutes above moderate activity could read between 33.6 and 75.7 minutes. The increase in size of the 95% limits of

agreement with intensity indicates that choice of epoch setting is important for assessment of all activity above light intensity. While some of these differences will be accounted for by inter-unit variation (Powell and Rowlands, 2004, Powell et al., 2003), the differences between time recorded by the two epoch settings is actually lower than that reported by Nilsson et al. (2002), where the same monitor was used to assess all epochs simultaneously so inter-instrument variation was not a factor.

As total physical activity increased, the likelihood of the underestimation of very hard activity when using a 60 s epoch also increased. With sedentary groups, or if activity intensity need only be classified as time spent in moderate intensity activity and above, a 60 s epoch setting may be adequate to obtain a full picture of activity. However, particularly in active populations, it is recommended that the 1 s epoch setting is used to maximise the accuracy of measurement of activity intensity. Problems may be encountered when assessing habitual physical activity (minimum 4 days assessment, Trost et al., 2000); at a 1 s epoch setting the memory capacity of the RT3 accelerometer is limited to 9 hours of monitoring, with the vector magnitude, and only 3 hours when using all three vectors. This does not allow an adequate measurement period, therefore it necessitates the download of activity data on a twice daily basis, compared to the 60 s epoch setting which allows up to 21 days of measurement (7 days if all three vectors are used and 21 days if only the vector magnitude is used).

Validity of monitors

The RT3 has been shown to be a valid tool for the assessment of physical activity in children and adults (Powell and Rowlands, 2004). However, inter-unit variability has been shown to exist (Powell and Rowlands, 2004; Powell et al., 2003) and therefore all monitors were electronically tested before inclusion in this study (Powell et al., 2003). Outliers (activity counts greater than two standard deviations

above or below the mean) were not included. The cut-off points used in the present study were developed across a range of laboratory activities including treadmill walking/running, passing a football, playing computer games and playing hopscotch (Rowlands et al., 2004). However, research has indicated that the relationship between accelerometer counts and metabolic demand differs depending on whether activity is assessed in the laboratory or the field (Nichols et al., 2000). However, as pointed out by Nilsson et al. (2002) the actual value of the cut-off points is not important as the epoch effect as activity intensity increases is the more influential factor here.

Conclusions

To our knowledge this is the first study to assess the time sampling effects of the RT3 accelerometer in the field. Epoch selection should be carefully considered when wishing to obtain a measure of vigorous intensity physical activity and greater. While a 60 s epoch allows longer data collection periods, it is likely that time spent in high intensity activity will be underestimated. This is particularly important when assessing the relationship between physical activity and bone health as high intensities of strain appear to be more beneficial to bone development than volume of activity (Parker, 1998). An increased memory capacity of the RT3 accelerometer would aid researchers wishing to quantify time spent at varying physical activity intensities.

Chapter 7

Study 5

Interactive effects of habitual physical activity and calcium intake on

bone density in boys and girls⁶

⁶ This study is in press with the Journal of Applied Physiology.

A version of this data was presented at Pediatric Work Physiology conference in Portugal, September 2003, by S.M. Powell and was awarded with the 'Young Researchers Award'.

This study was funded by the North Wales Research Committee: A Sub Committee of the North Wales Health and Social Care R & D Collaboration (see appendix D1). The authors would like to thank Sarah Stevens for assistance with data collection, and the children, parents and headteachers form Ysgol Faenol, Ysgol Felinheli, Ysgol Glan Cegin, Ysgol Hirael, Ysgol Llandegfan, Ysgol Penybryn, Ysgol Tregarth and Ysgol Y Borth, North Wales who volunteered to participate in this study.

Abstract

Purpose: To assess the interactive effects of habitual physical activity (total and vigorous intensity) and calcium intake on bone mineral content (BMC) in pre-pubertal boys and girls. Methods: Seventy-six children, aged 8-11 years, wore accelerometers for up to seven days to assess activity. Calcium intake was estimated by a four-day weighted food diary. BMC and areal density (BMD) were measured at the total body, proximal femur and femoral neck using dual energy X-ray absorptiometry. Moderated regression analyses were used to assess the contributions of physical activity (total and vigorous) and calcium intake to BMC, residualised for bone area and body mass (BMC_R). Results: Interactive effects of vigorous activity (≥ 6 METS) and calcium intake were found for the total body in boys ($b = 2.90 \times 10^{-3}$) and in girls ($b = 6.58 \times 10^{-3}$) ³), and at the proximal femur ($b = 9.87 \times 10^{-5}$) and femoral neck ($b = 2.29 \times 10^{-5}$) in boys only; BMC_R was high only if both vigorous activity and calcium intake were high. There were no interactive effects of total activity and calcium intake. Conclusions: This study provides evidence for the synergistic action of habitual vigorous activity and calcium intake on bone mass in children. Recommendations for optimising bone mass should reflect this synergism.

Introduction

Physical activity is widely acknowledged to have a positive effect on bone mass during growth (Bass et al., 1998; Parker, 1998; Turner and Robling, 2003). However, details regarding the magnitude and nature of this relationship are still unclear. Bone mass is increased by dynamic activity involving high strains and unusual strain distributions (Parker, 1998; Turner and Robling, 2003). This type of activity may be difficult to quantify using self-report measures of physical activity, especially in children as they lack the cognitive ability to recall details about their activity patterns (Livingstone, 1994; Rowlands et al., 1997). Conceptually, the ideal solution for the assessment of physical activity is the use of monitors that actually measure or track movement (Rowlands et al., 2000). Rowlands et al. (2002) used simple pedometry and found that habitual physical activity explained up to 14.3% of the variance in the size-adjusted bone mineral content (BMC) of the hip, in 8-11 year old children. However, pedometry records the total number of steps only and does not differentiate between intensities of activity.

Modern accelerometers can register accelerations and decelerations caused by bodily movements over several weeks. These accelerations and decelerations are integrated across user-defined epochs and stored for later download. Additionally, several published count cut-off values are available, allowing data to be expressed as minutes spent in varying intensities of physical activity (Eston et al., 1998; Rowlands et al., 1999). Therefore, temporal analysis of movement data is possible, permitting analysis of total physical activity, time accumulated at different intensities of activity, and the pattern of activity. The time resolution of the accelerometer is important when assessing activity relevant to bone density as this allows short periods of intense activity to be captured. For bone development, high intensities of strain to the musculoskeletal system are more important than the volume of activity (Parker, 1998). Other methods of assessing activity would likely miss these potentially important episodes. Consequently, physical activity assessed by one dimensional (uniaxial) accelerometry has been shown to account for 1.5% to 9% of the variance in size adjusted bone measures, in 4 to 6 year old children (Janz et al., 2001).

It is unclear whether physical activity or calcium intake is more important for bone mass accrual. Some research has shown a greater influence on bone mass from calcium supplementation than physical activity interventions (Ilick et al., 1998). Conversely, other studies have shown the contrary (Ruiz et al., 1995). It is well documented that a substantial percentage of children and adolescents, especially females, do not consume and absorb enough dietary calcium during the period of peak bone mass accretion (Chan, 1991; Weaver, 1996). Anderson (2000) concluded that, as few children consume optimal amounts of calcium, the beneficial effect of physical activity may dominate as a determinant of bone mass in early life.

To date, the statistical design of the majority of studies has considered calcium intake and physical activity to have independent effects on bone mass. However, potentially there is a synergistic action of calcium intake and physical activity, whereby both have to be high for an optimal effect on bone mass. Two studies have recently performed longitudinal analyses to address this question. Specker and Binkley (2003) used a 12-month, randomised, placebo-controlled intervention trial of activity (30 mins.d⁻¹ of fine or gross motor activities) and calcium (500 mg.d⁻¹ or placebo) in 178 children aged 3 to 5 years. After 12 months, there was little difference in leg BMC gain between gross and fine motor groups receiving the placebo (38.2 \pm 1.2 vs. 38.5 \pm 1.3 g, respectively), whereas when supplemented with calcium the gross motor group showed a 3.6 g higher BMC increase relative to the fine motor group (40.9 \pm 1.3 vs. 37.3 \pm 1.4 g respectively, p< 0.05). However, the gain in cortical area and thickness in the leg by the calcium-supplemented gross motor group was similar to the gain shown by the fine motor group receiving the placebo. Therefore, these results should be interpreted with caution.

Iuliano-Burns et al. (2003) found more consistent results following an 8.5month, randomised, placebo-controlled intervention trial of activity (low-impact or moderate-impact groups) and diet (calcium-fortified or non-fortified foods) in 66 preand early-pubertal girls. There was little difference in femoral BMC gain between the moderate- and low-impact exercise groups receiving the non-fortified foods ($25.4 \pm$ 2.2 cf. 26.6 ± 2.1g, respectively), whereas there was a 10.5g higher BMC gain in the moderate-impact exercise group relative to the low-impact exercise group receiving the calcium-fortified foods (31.7 ± 2.3 cf. $21.2 \pm 2.6g$, respectively p < 0.05).

However, such an activity intervention is not an option for most children. Therefore, it is important to explore whether there are interactive effects of naturally occurring physical activity and dietary calcium levels on bone mass. The aim of the present study was to assess the effects of habitual physical activity (total and vigorous intensity) and calcium intake on bone mineral content (BMC) in pre-pubertal boys and girls. We predicted that bone mass would be at its highest when both calcium intake and physical activity were high. Furthermore, we anticipated that this effect would be more evident for vigorous physical activity than for total physical activity.

Methods

Participants

Seventy-six children were recruited from eight primary schools in North Wales. The sample comprised 38 girls (mean \pm SD, age 9.0 \pm 1.0 yrs, mass 30.2 \pm 8.3 kg and height 130.4 \pm 7.0 cm) and 38 boys (age 9.1 \pm 0.7 yrs, mass 32.6 \pm 6.4 kg and

height 134.0 \pm 6.2 cm). Our pilot work suggested that we could expect a multiple correlation (with three independent variables) of approximately 0.35 ($R^2 = 0.12$). To detect an effect of this size, with a power of 0.80 and alpha of 0.05 would require a sample size of 34 children (Cohen, 1992). Boys and girls were to be analysed separately. All participants were healthy with no known diseases affecting bone metabolism.

Ethics approval was granted by the North Wales Health Authority Research Ethics Committee (see appendix D2). Written informed consent was obtained from all parents and verbal consent from all children (see appendix D3). Each participant and their parents were visited at home, where all procedures were explained (see appendix D4-5). A pack was administered containing an accelerometer, four food diary forms and a maturational status form, all with written instructions.

Anthropometric and maturational assessment

Height was measured to the nearest 0.1 cm and body mass to the nearest 0.1 kg using a free-standing Seca stadiometer and Seca scales (Seca AG, Reinach, Switzerland). The children self-reported their pubertal status, aided by parents, using Tanner stages of breast development for girls and pubic hair development for boys (Tanner, 1962). Girls were also questioned on their menstruation status.

Calcium Intake

Food intake was recorded continuously using a weighted food diary. Each child and their parent(s) were instructed how to keep the food diary and record the time and weight of all food and drink ingested. The diary was kept for three weekdays and one weekend day. Diaries were analysed for average daily calcium content in milligrams using commercially available software (Dietmaster, Version 4, Swift Computer Systems Ltd., Surrey, UK).

Physical Activity

Physical activity was measured using triaxial accelerometers (RT3, Stayhealthy Inc., Monrovia, CA, USA). Data were collected during term time between March and September in a single year. Each child wore an accelerometer for up to seven days. Data were scrutinised, and any day during which a monitor was evidently removed for more than 30 consecutive minutes was excluded from the data set. By this criterion, each child provided data for a minimum of four days (mean \pm SD = 5.2 \pm 0.9 days), as recommended by Trost et al. (2000). The accelerometer was worn on the right hip, from the time the child got up in the morning until bedtime. The accelerometer was initialised and downloaded via a computer interface and had no external controls that could be manipulated. The accelerometer was programmed to record activity counts in one-minute epochs.

The use of triaxial accelerometry to assess movement in more than one plane is potentially advantageous, as unusual strain distributions are particularly advantageous to BMD (Parker, 1998). However, there are inter-unit variability problems with the RT3 accelerometer along the anterioposterior (Y) and mediolateral (Z) planes of motion (Powell et al., 2003). Therefore, in the present study, activity was analysed using the vertical (X) plane of motion only. The vertical plane is considered to be the most important axis of measurement, given the well established relationship between impact-loading / weight bearing activity and bone mass accrual (Turner and Robling, 2003; Welten et al., 1994).

The mean daily activity count on the vertical axis (total activity), and mean times spent in very hard, hard, vigorous, moderate and low intensity activities were

used as the output measures for physical activity (Table 16). Additionally, the mean time spent at an intensity of vigorous and above was calculated (\geq vigorous activity).

Activity intensity	Accelerometer count	MET value	
Low	0-635	0-2.9	
Moderate	636 - 1645	3-5.9	
Vigorous	1646 - 2320	6-8.9	
Hard	2321 - 2825	9-11.9	
Very hard	2826+	12+	

Table 16. The relation of activity intensity to accelerometer counts and MET value.

Activity counts and MET equivalents taken from Rowlands et al. (2004).

Bone Mineral Content

Bone area (cm²), bone mineral content (BMC, g) and areal bone mineral density (BMD, g.cm⁻²) of the whole body and hip region, specifically the total proximal femur and femoral neck, were measured using dual energy X-ray absorptiometry (DXA, QDR-1500 Elite, Hologic Inc., Waltham, MA, USA, software version 7.10). Scans took place within two weeks of physical activity monitoring. The standard Hologic protocol for positioning was followed.

All scans were attended by the same two investigators (SMP or SS) and all scans were analysed by the same investigator (SMP) in our laboratory. Quality assurance was performed daily by scanning a spine phantom supplied by the manufacturer. The in-vivo precision error of DXA in our laboratory, expressed, as the coefficient of variation, is approximately 1.0% for the total proximal femur, 1.4% for the femoral neck and 0.5% for the whole body.

Statistical Analysis

Descriptive statistics were calculated for all variables. Independent *t*-tests were used to analyse gender differences in anthropometric and bone measures. A series of six independent *t*-tests were carried out to examine gender differences in time spent at the various activity intensities. Where necessary, degrees of freedom were adjusted owing to violation of the assumption of homogeneity of variance. The Bonferroni correction was used, reducing alpha to 0.008 (0.05/6), to allow for multiple tests of significance.

BMD is an areal density measurement (g.cm⁻²), i.e., BMC is divided by bone area. However, this does not adequately account for differing body sizes (Prentice et al., 1994). Therefore, BMC was regressed on bone area and body mass, and the residuals were saved to form a new variable, residualised BMC (BMC_R). Therefore, a child's BMC_R score represented the extent to which the child's actual BMC score exceeded or fell below what would be expected given the child's mass and bone area. Height was not included in this correction for body size as it did not account for any variance in BMC beyond that explained by bone area and body mass (see Prentice et al., 1994).

Multiple moderated regression analyses (Jaccard and Turrisi, 2003) were used to assess whether calcium intake moderated the relationship between total activity and total body residualised BMC (BMC_R-TB), proximal femur residualised BMC (BMC_R-PF) and femoral neck residualised BMC (BMC_R-FN). The independent variables were entered into the regression analysis in the following order: calcium intake, total activity, and the product of calcium intake and total activity (calcium intake * total activity). A significant product term would indicate an interactive effect of calcium intake and physical activity on BMC_R. To avoid the problem of multicollinearity, the independent variables (calcium intake and total activity) were centered prior to entry

into the analysis. Centering entailed subtracting the mean from each individual score; therefore the mean of the centered variable was zero. The product term was calculated from these centered variables. When interpreting the regression output the unstandardized solution was examined. To clarify the form of a significant interactive effect the relationship between total activity and BMC_R when calcium intake was medium (at its mean), high (1 SD above the mean) and low (1 SD below the mean), as described by Jaccard and Turrisi (2003), was graphed.

It should be noted that, as this is a product-term model, the regression coefficients for physical activity and calcium intake represent simple effects rather than main effects (Jaccard and Turrisi, 2003): the coefficient for physical activity estimates the effect of activity on BMC_R when calcium intake is at its mean (centered calcium intake is zero); and the coefficient for calcium intake estimates the effect of calcium on BMC_R when total activity is at its mean (centered total activity is zero).

The importance of high intensity activity was investigated by repeating the above analyses with \geq vigorous activity in place of total activity. All analyses were conducted separately for each gender. The possibility of multicollinearity was examined using variance inflation factor (VIF). In all cases, VIF was lower than 2.5 indicating that multicollinearity was not a cause for concern (Allison, 1999).

An alpha level of 0.05 was used for all statistical tests. SPSS version 11.0 for Windows (SPSS Inc., Chicago, IL.) was used for all statistical analyses.

Results

Descriptive data are shown in Table 17. Boys were taller than girls (P < 0.05). Boys' BMD was 6.5% higher than girls' at the total body ($t_{74} = -5.0$, P < 0.001), 13.3% higher at the proximal femur ($t_{74} = -4.9$, P < .001) and 14.0% higher at the femoral neck ($t_{74} = -5.7$, P < 0.001). Boys' total activity was 17.1% higher than girls' ($t_{60.7} = -3.07$, P < 0.005) and they spent 2.2% less time in low intensity activity ($t_{74} = 3.1$, P < 0.005), 46.0% more time in vigorous intensity activity ($t_{62.0} = -3.5$, P < 0.001), and 55.4% more time in \geq vigorous intensity activity ($t_{54.1} = -3.3$, P < 0.005).

The results of the regression analyses, examining the effects of calcium intake and physical activity on residualised BMC, are shown in Table 18. Results for total physical activity are presented in the upper half of Table 18. No calcium by total activity interactions were found at any site in boys or girls. There were simple effects for calcium at the total body in boys ($b = 6.26 \times 10^{-2}$, SE = 0.03) and girls ($b = 9.34 \times 10^{-2}$, SE = 0.04), but no simple effects of total activity at the total body in boys or girls. In boys only, there were simple effects of calcium intake and total activity at the proximal femur ($b = 2.05 \times 10^{-3}$, SE = 0.00; $b = 1.05 \times 10^{-5}$, SE = 0.00, respectively) and femoral neck ($b = 4.32 \times 10^{-9}$, SE = 0.00; $b = 2.05 \times 10^{-6}$, SE = 0.00, respectively).

Results for \geq vigorous activity are presented in the lower half of Table 18. There was a calcium by \geq vigorous activity interaction at the total body in boys ($b = 2.9 \times 10^{-3}$, SE = 0.00) and girls ($b = 6.58 \times 10^{-3}$, SE = 0.00). In boys only, interactive effects were also present at the proximal femur ($b = 9.87 \times 10^{-5}$, SE = 0.00) and the femoral neck ($b = 2.29 \times 10^{-5}$, SE = 0.00). The interactive effects are depicted in Figures 21-24. Each graph shows the relationship between \geq vigorous activity and BMC_R for three different levels of calcium intake: low (1 SD below the mean), medium (at the mean), and high (1 SD above the mean). These graphs indicate that, where an interactive effect was present, BMC_R was only high if both calcium and \geq vigorous activity were high, with no benefits from one variable alone being high. This was the case at the total body in boys and girls (Figures 21 and 22) and at the proximal femur and femoral neck in boys only (Figures 23 and 24). Furthermore, in boys \geq vigorous activity had a positive effect at the total body, proximal femur and femoral neck if calcium intake was at its mean or higher (Figures 21, 23 and 24), whereas in girls \geq vigorous activity only had a positive effect at the total body if calcium intake was 1 SD above the mean (Figure 22).

	Boys $(n = 38)$	Girls (<i>n</i> = 38)
Age (years)	9.1 ± 0.7	9.0 ± 1.0
Mass (kg)	32.6 ± 6.4	30.2 ± 8.3
Height (cm)	134.0 ± 6.2*	130.4 ± 7.0
Calcium intake (mg.d ⁻¹)	762.9 ± 310.1	672.6 ± 177.6
Pubertal stage	1.1 ± 0.3	1.2 ± 0.4
BMC-TB (g)	$1033.3 \pm 186.4^{\dagger}$	901.7 ± 189.2
BMD-TB (g.cm ⁻²)	$0.870 \pm 0.050^{+}$	0.817 ± 0.042
BMC _R -TB	22.937 <u>+</u> 46.194	-16.751 <u>+</u> 42.778
BMC-PF (g)	$13.8 \pm 3.1^{\dagger}$	11.8 ± 2.9
BMD-PF (g.cm ⁻²)	$0.741 \pm 0.084^{\dagger}$	0.654 ± 0.070
BMC _R -PF	0.748 <u>+</u> 1.455	-0.483 <u>+</u> 1.017
BMC-FN (g)	$3.4 \pm 0.5^{+}$	2.8 ± 0.4
BMD-FN (g.cm ⁻²)	$0.701 \pm 0.073^{\dagger}$	0.615 ± 0.059
BMC _R -FN	0.192 <u>+</u> 0.327	-0.126 <u>+</u> 0.239
Total activity (counts)	315367.3 ± 79301.2 [†]	269251.4 ± 47794.2
Low intensity activity (mins)	$1266.9 \pm 44.0^{+}$	1295.2 ± 35.5
Moderate intensity activity (mins)	135.8 ± 42.7	118.6 ± 27.9
Vigorous intensity activity (mins)	$28.9 \pm 13.4^{+}$	19.8 ± 8.3
Hard intensity activity (mins)	8.0 ± 7.2	4.9 ± 3.1
Very hard intensity activity (mins)	4.7 ± 6.8	2.0 ± 1.8
\geq vigorous intensity activity (mins)	$41.5 \pm 25.2^{\dagger}$	26.7 ± 12.4

Table 17. Descriptive statistics (mean ± SD).

BMC = Bone mineral content, BMC_R = residualised bone mineral content, BMD = bone mineral density, TB = total body, PF = proximal femur, FN = femoral neck. * P < 0.05, † P < 0.008 for gender difference.

Site	Predictor variables in	Boys		Girls	
	order of entry				
<u> </u>		R^2 change	B(SE)	R ² change	b(SE)
		Total A	ctivity	<u></u>	
BMC _R -TB	Calcium	0.08	6.26x10 ⁻² (0.03)*	0.18*	9.34x10 ⁻² (0.04)*
	Total activity	0.06	2.03x10 ⁻⁴ (0.00)	0.04	1.82x10 ⁻⁴ (0.00)
	Calcium x total activity	0.01	3.95x10 ⁻⁷ (0.00)	0.01	5.51x10 ⁻⁷ (0.00)
BMC _R -PF	Calcium	0.03	2.05x10 ⁻³ (0.00)*	0.04	1.23x10 ⁻³ (0.00)
	Total activity	0.12*	1.05x10 ⁻⁵ (0.00)*	0.00	1.17x10 ⁻⁶ (0.00)
	Calcium x total activity	0.07	2.89x10 ⁻⁸ (0.00)	0.00	-6.94x10 ⁻⁹ (0.00)
BMC _R -FN	Calcium	0.03	4.32x10 ⁻⁴ (0.00)*	0.03	2.84x10 ⁻⁴ (0.00)
	Total activity	0.10*	2.05x10 ⁻⁶ (0.00)*	0.00	-9.41x10 ⁻⁸ (0.00)
	Calcium x total activity	0.04	5.14x10 ⁻⁹ (0.00)	0.01	-2.96x10 ⁻⁹ (0.00)
		Vigorous	Activity		
BMC _R -TB	Calcium	0.08	7.81x10 ⁻² (0.03)*	0.18*	9.39x10 ⁻² (0.04)*
	Vigorous Activity	0.11*	9.1x10 ⁻¹ (0.31)*	0.03	5.80x10 ⁻¹ (0.50)
	Calcium x vig. activity	0.09*	2.90x10 ⁻³ (0.00)*	0.09*	6.58x10 ⁻³ (0.00)*
BMC _R -PF	Calcium	0.03	2.12x10 ⁻³ (0.00)*	0.04	1.20x10 ⁻³ (0.00)
	Vigorous Activity	0.23*	3.77x10 ⁻² (0.01)*	0.01	-8.76x10 ⁻³ (0.01)
	Calcium x vig. activity	0.10*	9.87x10 ⁻⁵ (0.00)*	0.00	9.60x10 ⁻⁶ (0.00)
BMC _R -FN	Calcium	0.03	5.01x10 ⁻⁴ (0.00)*	0.03	2.75x10 ⁻⁴ (0.00)
	Vigorous Activity	0.21*	8.36x10 ⁻³ (0.00)*	0.03	-3.3x10 ⁻³ (0.00)
	Calcium x vig. activity	0.11*	2.29x10 ⁻⁵ (0.00)*	0.01	-1.15x10 ⁻⁵ (0.00)

Table 18. Multiple moderated regression analysis results.

Note. BMC = Bone Mineral Content. BMC_R = Residualised Bone Mineral Content. TB = Total Body. PF = Proximal Femur. FN = Femoral Neck. b = regression coefficient from final equation. R^2 change = increment in variance explained when entering predictor. Calcium intake and physical activity scores were centred prior to entry into the analysis. Interaction terms were calculated from the centered scores. * P < 0.05



Figure 21. Vigorous activity x calcium interaction on residualised bone mineral content at the total body in boys.


Figure 22. Vigorous activity x calcium interaction on residualised bone mineral content at the total body in girls.



Figure 23. Vigorous activity x calcium interaction on residualised bone mineral content at the proximal femur in boys.



Figure 24. Vigorous activity x calcium interaction on residualised bone mineral content at the femoral neck in boys.

Discussion

As hypothesized, BMC_R was highest when both vigorous activity and calcium intake were high. This was the case for the total body, proximal femur and femoral neck in boys, but only for the total body in girls. These interactive effects were not apparent for total physical activity, although there were simple effects for total activity at the hip in boys, calcium at the total body and hip in boys and at the total body only in girls.

These results extend the findings of Specker and Binkley (2003) and Iuliano-Burns et al. (2003) who both reported that an exercise intervention resulted in gains in bone mass at the leg or hip only when calcium intake was also supplemented. It appears the same effect is present with habitual levels of vigorous activity and calcium intake in boys. However, in contrast to the current study, neither intervention study found an interactive effect of exercise and calcium at the total body.

It is surprising that few studies have addressed the interactive effect of calcium intake and physical activity on bone, as calcium appears necessary for exercise to have an optimal bone stimulating action (Branca et al., 2001). Conclusions from studies that have addressed only the main effects of calcium intake or physical activity on bone may be misleading. For example, in a non-moderated regression model the main effect of physical activity on bone would be assessed across all levels of calcium intake (Jaccard and Turrisi, 2003). This would dilute any effect of physical activity on bone that only occurred at relatively high calcium intakes.

The importance of vigorous activity is not surprising as high intensities of strain to the musculoskeletal system appear to be more important than the volume of activity to bone development (Parker, 1998). Our results support Janz et al. (2001), who reported higher positive correlations of hip BMC with vigorous activity (r = 0.25 in boys and r = 0.28 in girls, P < 0.05) than with total physical activity (r = 0.20 in boys and r = 0.25 in girls, P < 0.05) in 4-6 year old American children. As well as high intensity activity, high frequency of activity is important for the effective application of mechanical forces which promote osteogenesis (Turner and Robling, 2003). The required mechanical load necessary to initiate new bone formation decreases as the loading frequency increases (Turner and Robling, 2003). This could contribute to the presence of a relationship between vigorous activity and residualised bone mass at the hip in the current study for boys, though not girls. The significantly lower number of minutes spent in vigorous activity in girls may indicate a lower frequency of activity.

Although not statistically significant, there was a trend for a higher calcium intake in boys (762.9 \pm 310.1 mg.d⁻¹) than girls (672.6 \pm 177.6 mg.d⁻¹). In fact, high calcium intake in girls (1 SD above the mean = 850.2 mg.d⁻¹) was similar to the mean score for boys. This is important as the interactions at the total body showed that vigorous activity had a beneficial effect on bone mass when calcium intake was at its mean or above in boys, but only if calcium intake was high (1 SD above the mean) in girls. Therefore, it is possible that there is a threshold calcium intake of 700-800 mg.d⁻¹ before vigorous activity impacts significantly on bone. This value is similar to the recommended dietary intake for 7-10 year olds (RDA) of 800 mg.d⁻¹ in the USA, but higher than the recommended nutrient intake (RNI) of 550 mg.d⁻¹ in the UK. It is notable that the *b* coefficients for vigorous activity when calcium intake is high are very similar for boys and girls (boys: *b* = 1.806; girls: *b* = 1.751). This indicates that, when calcium intake is high, the nature of the relationship between vigorous activity and BMC_R-TB is similar for boys and girls.

Assuming calcium intake was 700-800 mg.d⁻¹, 40 minutes per day of vigorous activity in boys, and 25 minutes of vigorous activity in girls, was associated with increased BMC_R. This quantity of vigorous activity supports the conclusions of Janz et al. (2001) who recommended that, to increase BMC at the hip and spine in 4-6 yr olds by 2-3% of the mean, vigorous activity needed to be increased by 10 - 40 minutes in boys and 35 minutes in girls. The effect in the present study is stronger, probably mainly due to no consideration of calcium intake, either in isolation or as an interactive effect with activity by Janz and colleagues.

The use of accelerometers set at a one minute epoch may have resulted in an underestimation of time spent in vigorous activity (Nilsson et al., 2002). This may have impacted on the size of the detected relationships between vigorous activity and BMC_R . Bailey et al. (1995) observed that children engaged in very short bursts of

intense physical activity interspersed with varying intervals of low and moderate intensity. The median duration of high intensity activities was found to be only 3 seconds with 95% lasting less than 15 seconds. Therefore, in future, it would be more appropriate to use a one second epoch to capture a more precise picture of vigorous physical activity in children. However, the memory capacity of the accelerometers precludes the use of this epoch setting for longer than 7 hours at present.

In conclusion, the evidence suggests that calcium intake and vigorous activity have a synergistic effect on bone. Results indicate that 8-11 year old children should participate in approximately 25-40 minutes per day of vigorous activity (\geq 6 METS) and dietary calcium intake should be 700-800mg.d⁻¹ for a positive impact on bone. It is notable that this calcium intake is higher than the current UK RNI of 550 mg.d⁻¹. However, it must be acknowledged that weighted food diaries are potentially prone to reporter bias and increased food awareness whilst recording dietary intake.

Additionally, current recommendations for children's physical activity recommend 60 minutes of moderate activity (\geq 3 METS) per day (Biddle et al., 1998). Further research is needed to investigate whether there are grounds to adapt these recommendations to optimise bone health in children, particularly as it appears that activity needs to be of a vigorous nature to stimulate an osteogenic response. This study was cross-sectional in nature limiting the conclusions that can be drawn; the effect of habitual physical activity and dietary calcium should also be investigated in a longitudinal study to allow the effect of changes in calcium intake and physical activity on bone mass in children, across different pubertal stages, to be examined.

Chapter 8

Conclusions

Main findings

This thesis has provided evidence for the synergistic action of habitual vigorous activity and calcium intake on bone mass in children, and established the RT3 accelerometer as an acceptable tool for the objective assessment of physical activity. Further research is necessary to establish whether, in the United Kingdom, a review of the guidelines offered for physical activity and calcium intake in children is needed.

This first study of the thesis (chapter 3) used pedometry to assess the relationship between habitual physical activity and bone mass in children. Steps per day were significantly related to the BMD and size-adjusted BMC of the total proximal femur and femoral neck in 8 to 11 year old children. This relationship persisted after accounting for calcium intake. The use of an objective measure of physical activity reduced the measurement error associated with physical activity assessment (Jones and Dwyer, 1998). However, pedometry provides only a total measure of activity and the use of accelerometry was recommended to gain a temporal picture of activity, in terms of intensity, frequency and duration of motion.

The next three empirical studies (chapters 4, 5 and 6) critically examined the RT3 accelerometer, as a means for objective physical activity assessment over three orthogonal planes including, intensity, frequency and durational data. Study two (Chapter 4) examined the inter-monitor technical variability of the RT3 accelerometer in a tightly controlled laboratory setting. At a low frequency (2.1Hz), reflective of most human motion, there were no differences between the three orthogonal axes.

However, at higher frequencies (5.1 and 10.2 Hz) the anterioposterior (Y) axis consistently read higher than both the vertical (X) and mediolateral (Z) axes. Intraclass correlation coefficients were high for the total sample and for individual axes (r = 0.99), reflecting the strong relationship between axes across all frequencies. However, intra- and inter-monitor coefficient of variations reflected the greater variability within and between the RT3s at low compared to high frequencies. Intraand inter-monitor comparisons compare favourably with published research on the CSA uniaxial accelerometer (Metcalf et al., 2002). It was recommended that reliability and variability of the RT3s be assessed over larger amplitudes of motion to generate results for more direct comparison with human physical activity assessment.

Study three (chapter 5) investigated the reliability and inter-monitor variability of the RT3 accelerometers, along each orthogonal axis of motion, over a range of typical standardised physical activities, in a controlled laboratory setting. Individual RT3 monitors were reliable over trials (with the exception of one monitor on the Y axis) and largely differentiated between low intensities of activities. However, differentiation decreased as activity intensity increased. Considerable inter-monitor differences within activities were also apparent on all axes, with the X axis of motion revealing the least variability between monitors.

Study four (chapter 6) examined the effect of epoch selection (60 second and 1 second) of the RT3 accelerometer on recorded habitual physical activity (total and minutes spent at varying intensities) in children. Vigorous intensity activity was significantly overestimated and very hard intensity activity was significantly underestimated when a 60 second epoch was used, although no epoch effects were evident for total physical activity or time spent in light, moderate or hard activity. Based on these three studies, it was recommended that all future research using the RT3s should perform *a priori* trials to identify any outlying monitors and to assess the

intra- and inter-monitor variability and reliability before use either in laboratory or field settings. If vigorous or high intensity activity is to be assessed a one second epoch should be used.

The last empirical study (chapter 7) used the RT3 accelerometer to objectively measure habitual physical activity (total and vigorous intensity) and examine the relationship between physical activity, calcium intake, and BMC (hip and total body) in pre-pubertal boys and girls. The use of multiple moderated regression analyses allowed the examination of calcium intake as a moderator of the relationship between physical activity and bone mass. Results provided evidence for a synergistic action between calcium intake and vigorous physical activity on bone mass. As hypothesized, BMC was at its highest when both activity and calcium intake were high, at all sites in boys, and the total body only in girls. Therefore, conclusions from previous studies that have considered calcium intake and physical activity as separate entities may be misleading. The results indicate current guidelines for physical activity and calcium intake may be too low for optimal bone health. This study calls for further research to support the possibility of a need for a re-assessment of the current guidelines for physical activity and calcium intake.

Methodological Issues

Internal validity

The validity with which statements can be made about whether there is a causal relationship from one variable to another, in the form in which variables were manipulated or measured, can be considered in each study (Vincent, 1999). Two measures which may reduce internal validity are instrument error and investigator error.

Studies 1 and 5

All children involved in these studies were requested to maintain normal daily activities, and dietary intake, and to do nothing out of the ordinary during the measurement period. After consent was given for the study, the children's parents or guardians were visited to discuss all procedures relating to the pedometer or accelerometer and dietary sheets. In study 1, parents sealed and placed the pedometer on the child's hip at the start of each day recording the time. At the end of the day the parent removed the pedometer, recorded the time and activity count and reset the pedometer for the next day. In study 5, the accelerometers were sealed throughout and so were merely placed on and removed from the child's hip at the start and end of each day. Therefore, internal validity rose from study 1 to study 5 as parents' requirements were reduced and so potential bias of recording activity counts, start and end time was removed. Children were blinded to the purpose of the study and given an acclimatisation period of two days. Therefore, reactivity was minimised. Although measures have been taken to reduce reactivity it cannot be ruled out that children may have behaved outside of their normal activity and dietary patterns thereby, possibly influencing the results.

In both studies, parents were responsible for the weighing of all food and fluid the child ingested over the four-day measurement period. The use of weighted food diaries reduced the possible bias created by the use of food frequency or self-report diaries, and permitted the accurate analysis of calcium intake within the habitual diet. It is recognised, however that investigator error may have been present in the form of parental reactivity to the detailed description of the diets they provide for the children. Instrument error was minimised as all food diaries were analysed by the same investigator using the same software.

All procedures for the assessment of BMD and BMC were explained to the children and parents at the location of measurement for studies 1 and 5. All questions were answered and all children were happy and relaxed throughout the analysis. All scans took place within one week of activity assessment. The standard Hologic protocol for positioning was maintained. All scans were conducted by one of two investigators but analysed by the same investigator in our laboratory, reducing potential investigator bias. Quality assurance and calibration were performed daily by scanning a spine phantom supplied by the manufacturer, minimising instrument error. The in-vivo precision error of dual energy x-ray absorptiometry (DXA) in our laboratory is approximately 1.0% for the total proximal femur and 0.5% for the total body. For a more detailed discussion of the internal validity of DXA see appendix (B).

It must also be acknowledged that due to the findings of study 4, there may have been a potential underestimation of \geq vigorous activity in study 5 as the 60 s epoch was used. However, recognition of the underestimation of \geq vigorous activity would only serve to increase the validity of the results.

Studies 2, 3 and 4

The results of study 1 called for intensity, frequency and durational measures of habitual physical activity and therefore, the RT3 accelerometer was placed under rigorous tests in the laboratory and field to ensure internal validity before use in study 5.

In study 2, internal validity can be assured as rigorous procedures were followed. Before each testing session apparatus was run for eight minutes at each frequency used to stabilise temperature and friction characteristics. Each RT3 was mounted securely in the test gig which was screwed directly to the aluminium vibration table, even at the highest test frequency this can be regarded as a rigid

structure. The vibration table which each RT3 was securely mounted, was driven by a moving coil armature via a power amplifier. The amplifier input was programmed accurately and reliably (pilot tests) to vibrate the RT3s at each of the three frequencies continuously with no interruptions. To reinforce accuracy of the sinusoidal wave input, a Kyowa (AS-10B) strain gauge accelerometer was mounted on the vibration plate also and the output was monitored on an oscilloscope at intervals with an estimated reading accuracy of $\pm 2\%$ and observed to be unchanged (for more detailed descriptions of procedures see chapter 4). Therefore, it can be assured minimal investigator error was involved and instrument procedures were rigorous to prevent error.

In study 3, one participant who was familiar with all activities performed all trials, and so no familiarisation was needed and learning effects were not evident. The RT3s were divided into two groups which, were securely taped together and moved as a whole, minimising instrument error. Each block measured $68 \times 48 \times 72$ mm and varied no more than \pm 36 mm from the centre line of the body. Ideally, all activities should have been presented in a random order to eliminate any effects due to the order of activity. This was not the case in this study and is recognised as a limitation. Activities were performed in a sequential order: rest, sit-stand, walking to running with two days in-between trials to allow for recovery and prevent fatigue effects. The first and last minute of each 12 minute bout was deleted to prevent sampling errors.

In study 4, the sub-sample of RT3s was randomly selected. From the three RT3s worn, two were taped securely together and attached to a belt, secured around each child's waist underneath their clothing to minimise awareness. It must be noted the intra- and inter-monitor variability demonstrated in studies 2 and 3 may have led to some error in the assessment of physical activity in study 5. However, this was

minimised by assessing the vertical vector (X) only where there was least intra- and inter-monitor variability.

Statistical Conclusion Validity

The power of the statistical tests of significance was considered in the planning of each study. By calculating the statistical power of the study *a priori* where possible (studies 1, 2 and 5), the assurance of data sensitive enough to make reasonable conclusions was sought.

Study 1

Pilot work suggested a correlation of approximately 0.35-0.40 (R² = 0.12-0.16) could be expected. To detect this effect size, with a power of 0.80 and an alpha level of 0.05, required a sample size of between 50 and 60 children in each group. As the sample size obtained was only 57 children, the analysis of genders separately would have led to an increased risk of Type II error (the failure to detect an effect that may have been present). Therefore, the genders were analysed together. This is recognised as a limitation as there remains the possibility that gender may act as a confounding variable (see chapter 3).

Study 2

In this controlled laboratory test it was hypothesized there would be a strong relationship between monitors and between axes at each frequency. Previous research with the CSA electronic accelerometer has shown intra-class correlations (ICC) greater than 0.8 between monitors tested at both fast and medium speeds and interinstrument repeatability greater than 0.77. The sample size necessary to detect this

effect size with a power of 0.80 and an alpha level of 0.05 is 11 (Cohen, 1992). The sample size in this study was 23 accelerometers.

Studies 3 and 4

These laboratory and field based studies are relatively novel which made it very difficult to predict the sample size needed as no predicted effects sizes were available. Additionally, no work has yet been completed to allow predictions for two or three way repeated measures designs (Stevens, 1996). However, in Study 3 a repeated measures design was used which increases statistical power (Stevens, 1996). In study 4, to detect a large effect size between two groups (60 s and 1 s epochs) with an alpha level of 0.05, approximately 26 subjects were needed (Cohen, 1992). However, the effects sizes found by Nilsson et al. (2002) were substantially larger than Cohen's large effect size. Therefore, our sample of 25 was more than adequate to see a power of 0.8.

Study 5

The results of pilot work and study one suggested a multiple correlation (with three independent variables) of 0.35 ($R^2 = 0.12$) could be expected. To detect this effect size, with a power of 0.80 and an alpha level of 0.05, required a minimum sample size of 34 children in each group (Cohen, 1992). A sample of 76, 38 boys and 38 girls, was obtained permitting the analyses separately by gender.

External validity

This refers to the ability to which results can be generalised to the population subjects were drawn from and results applied to the 'real world' (Vincent, 1999).

Study 1 and 5 in this thesis considered only one age group; eight to 11 year old pre-pubertal children. It is recognised that this limits the generalisability of the results where older and younger children are concerned. Due to ethical limitations of DXA use with older menstruating females, time, monetary and sample size constraints it would not have been possible to sample children from a wider age range and pubertal stage. In both studies, male and female volunteers were taken from eight primary schools in the local area, some with higher free school meal status than others, therefore, to a certain extent results are generalisable across social economic status and fully across gender. However, as in the majority of activity studies which require active participation all subjects were volunteers and therefore, it was impossible to obtain a random sample. Mean height and body mass of all children were within normal values for their respective populations (Tanner and Whitehouse, 1984) therefore, it appears the sample was representative. External validity was enhanced by gauging habitual physical activity and not influencing behaviour in any way.

Study 2 from this thesis used all 23 RT3 accelerometers bought from the company to assess technical variability. External validity was limited in this study as all procedures were in a rigorously controlled laboratory setting with RT3s being tested by frequency vibration incongruent to human movement. External validity was enhanced in study 3 by including physical activities which were considered representative of the types of movement children make during normal behaviour and measured on a human rather than by a machine. However, external validity could have been further enhanced by using a child participant rather than an adult. Again external validity was increased in study 4 by using a field setting and measuring habitual physical activity.

Implications and Future Research

This thesis has provided evidence supporting relationships between physical activity, calcium intake and bone mass in children. Through previous cross-sectional and retrospective analysis of athletes, research has shown a direct link between physical training and bone mass in the young (Bass et al., 1998; Bennell et al., 2000; Kontulainen et al., 2001; Lehtonen-Veromaa et al., 2000), which has been supported by longitudinal activity interventions in both boys and girls (Bradney et al., 1998; Fuchs et al., 2001). Study 1 adds to this body of research by extending the results to objectively assessed habitual levels of physical activity. This makes the results applicable to normal populations of children as well as the sporting elite and those with access to an activity intervention. Estimates were made regarding the amount of additional activity needed to bridge the gap between those with low compared to high activity (approximately 30 minutes). However, there were limitations to this study outlined in chapter 3, namely the restrictive effects sample size had on the separate analysis of boys and girls data and the use of pedometry, limiting habitual activity data to a counts per day total. Therefore, it was not possible to assess whether children met national guidelines for habitual physical activity.

Without an accurate measure of physical activity, relationships between activity and health indices are difficult to quantify. The RT3 tri-axial accelerometer has an advantage over the validated CSA uni-axial accelerometer, namely the ability to record frequency, intensity and durational information across three planes of motion as opposed to one, adding to the research potential. However, the results of studies 2 and 3 raised concerns regarding intra- and inter-monitor variability. Therefore, recommendations are made to other research groups using RT3s that strict quality control procedures were necessary before use to allow for confidence in the data obtained. Concern over intra- and inter-monitor variability along the Y and Z axes

limited the use to the X axis of the RT3 only in this sample removing the benefits of its tri-axial nature.

The results from studies 2, 3 and 4 have implications for future use and research. When using a number of activity monitors in a single study it is important the inter-monitor variability is minimal, if not, comparison of activity levels between participants is not possible. Acceptable parameters of variability depend on the nature of the research and should be outlined by laboratories using RT3 monitors. Future research is needed to clarify the technical reliability of the RT3s using apparatus with a larger field of movement to generate results for more direct comparison with human movement. If this research was carried out with the addition of repeatability testing, with multiple samples of RT3s, a standard procedure could be compiled allowing cross-comparison of samples, and recommendations regarding how often the calibration of RT3s should be checked. The publication of standard variability of axes at given frequencies would also allow the direct adjustment of data obtained. Additionally, it would be beneficial for the accelerometers to have a larger memory capacity to aid the use of the 1s epoch over a longer period of time, ideally seven days, to quantify time spent at varying intensities of activity accurately.

Despite these limitations and recommendations, the X (vertical) axis of motion was reliable over time, differentiated well between activities and elicited the least variability within each activity tested. Coupled with the fact that the RT3 accelerometer is user friendly and portable in terms of set-up and download it may be considered a reliable tool at least as a uniaxial monitor, recording data along a vertical axis of motion only. The vertical plane is considered to be the most important axis of measurement, given the well established relationship between impact-loading / weight bearing activity and bone mass accrual (Turner and Robling, 2003; Welten et al.,

1994), therefore, the RT3 accelerometer was used to measure physical activity along the vertical plane in study 5.

As hypothesized there was an interactive effect of habitual vigorous physical activity and calcium intake on bone mass in boys and girls. These results support previous findings of intervention studies which demonstrated bone mass benefited from the supplementation of both physical activity and calcium intake, but from neither intervention alone (Specker and Binkley, 2003; Iuliano-Burns et al., 2003). This study employed a statistical design which demonstrated the moderating effects calcium intake had on the relationship between physical activity and bone mass. Conclusions from previous research to date that have only assessed the main effects of calcium intake or physical activity on bone may be misleading. Recommendations from this research are that 8-11 year old children should participate in approximately 25-40 minutes per day of vigorous activity and have a dietary calcium intake of at least 700-800mg.d⁻¹ for a positive impact on bone. This would extend the current recommendation for children's physical activity of 60 minutes of moderate activity and substantially exceeds than the UK RNI for calcium intake of 550 mg.d⁻¹.

This study was cross-sectional in design limiting the conclusions that may be drawn. Further research of a longitudinal nature is necessary to confirm the possible need in the United Kingdom for a re-analysis of the guidelines offered to children in terms of bone health. Appropriate statistical designs should be employed to prevent misleading results and the search for valid and reliable tools for activity measurement across all three planes of human motion should be continued. It is likely that multiple measurements of physical activity will help elucidate relationships between physical activity and different aspects of health further.

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PAGE MISSING IN ORIGINAL

- 1) O V Jones Bursary (funding)
- 2) Ethics approval
- 3) Informed consent forms



Clinical Pathology Accredited

Ein Cyf/Our Ref: KDG/EMS

Estyniad/Extension 4259 or 4275

20 June, 2000

Miss Sarah Powell School of Sport Health and Exercise Sciences University of Wales Bangor

Dear Sarah

Re: Mr O V Jones Summer Bursary

I am delighted to inform you that you have been awarded the above bursary for the year 2000. The award is for £1500 and we will leave it up to your department to invoice us when required. I would be pleased if you could arrange for the invoice to be made out to the North Wales Health Authority and sent to Liz James.

I am sure the research will be successful and one of the conditions of the bursary is that we receive a report at the end of the project.

Once again, congratulations.

Best wishes

Yours sincerely

DR K D GRIFFITHS BSc., PhD., CChem, MRSC., CONSULTANT BIOCHEMIST/CHAIRMAN, R & D

cc Dr Roger Eston, Head of School Dr Ann Rowlands Pwyllgor Moeseg Ymchwil Awdurdod Iechyd Gogledd Cymru (Is-bwyllgorau'r Gorllewin, y Canol a'r Dwyrain)

> North Wales Health Authority Research Ethics Committee (West, Central & East sub-committees)

IS-BWYLLGOR Y GORLLEWIN WEST SUB-COMMITTEE

<pre>ttm/Tel: (U1248) 384 877 (llinell uniongyrchol/direct line)</pre>	Uned Cefnogi Rheolaeth Glinigol/
Ffacs/Faz : (01248) 370 629	Clinical Governance Support Unit
Llyth-cl/E-mail : liz.james@nww-tr.wales.nhs.uk	Ysbyty Gwynedd
•	Bangor
	Gwynedd LL57 2PW

Dr A Rowlands SSHES
•••••
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•••••••

Dear Ann

Acknowledgement of Receipt

I confirm that the North Wales Health Authority Research Ethics Committee (West) has received and reviewed the following:

"95/71: bone mass in girls, aged &-10 years" subsample obidy ortension as per your

e-mail 7.6.02 - approved.

Accepted/Approved on behalf of the Committee Signed

Dr Patricia Barry, Chairman

Date 8.7.02

nddirledolaeth GIG Gogledd Orllewin Cymru, Yabyty Gwynedd, Bargor, Gwynedd LL57 2PW Pflin/Tei: 01248 384384 • Fface/Fer: 01248 370829

Chairman/Cadelrydd - Dr P Barry





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Annwyl Riant/Gwarcheidwad,

Ξ.

Rydymun cynnal ymchwil I'r berthynas rhwng gweithgarwch corfforol a dwyster mwyn esgyrn mewn plant ym Mhrifysgol Cymru Bangor. Rydym am asesu os yw lefel gweithgarwch corfforol plant yn cael effaith fuddiol ar iechyd y sgerbwd. Mae hyn yn bwysig, oherwydd gall gryfhau mas yr esgyrn pan yn ifanc warhardd toriadau a mal yr esgyrn yn hwyrach mewn bywyd.

Rydym yn chwilio am ugain o wirfoddolwyr benyw rhwng 8 a 10 mlwudd oed. Bydd gofyn i'r gwirfoddolwyr yma wisgo peiriant monitor gweithgarwch, sydd yn fychain iawn (tua maint bocs fatys, sydd yn cael ei clipio I felt neu band wast) am saith diwrnod. Ar ddiwedd y wythnos bydd taldra, pwysau a dwyster esgyrn y plant yn cael ei fesur. Bydd angen ffurflen yn dyn odi statws aeddfedrwydd eich plentyn. Bydd dwyster mwyn yr asgwrn yn cael ei fesur gan ddefnyddio periant DEXA (dual energy x-ray absorptiometry). Mae'r dechneg hon yn gofyn ir plentyn orwedd ar y bwrdd tra bydd y glyn yn cael ei scanio, (dim ond 6 munud bydd y dechneg hon yn gymeryd ac mae'n hollol saff a rhydd o boen). Mae'r peiriant DEXA yn defnyddio pelydrau-x i wneud y mesuriadau. Mae'r pelydrau yma yn isel iawn o gymharu a gweithredau pelydr-x eraill, e.e y rhai a ddefnyddir gan y deintydd. Mae hefyd yn is na'r pelydrau rydym yn eu derbyn yn yr amgylchedd bob dydd. O'n profiad ni mae plant yn dangos diddordeb ac yn derbyn pleser mawr o gymeryd rhan mewn astudiaeth fel hon. Bydd y wybodeath am lefel gweithgarwch eich plentyn a chanlyniadau dwyster yr asgwrn ar gael i chi ar ol chwbwlhaeu'r astudiaeth. Gall y wybodaeth yma gael ei gyhoeddi ond byddwch yn gwirfoddoli yn ddi enw.

Bydd tranfidiaeth ir Brifysrol ac yn ol yn cael ei drefnu, ac mae croeso i chi ddod gydach plentyn. Os oes gan eich plentyn ddiddordeb mewn gwirfoddoli ar gyfer yr astudiaethyma, ac byddwch yn cytuno, a wnewch chi arwyddo'r ffurfirn isod os gwerthfawrogi cyfraniad eich plentyn yn fawr.

Yr eiddoch yn gywir,

Sarah Powell

Rwy'n tystio fod fy mhlentyn yn cymeryd rhan yn y prosiect yma o'i ewyllys ei hyn ac gallwn I neu fy mhlentyn

derfynnu ein cyfranniad ar unrhyw bryd.		
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Cyfeiriad_

Appendix B: Dual energy X-ray absorptiometry

Appendix B: Dual Energy X-ray Absorptiometry

Dual energy X-ray absorptiometry (DXA) was introduced in the 1980's as the use of dual photon absorptiometry (DPA) declined. It is now the most widely used bone mineral density (BMD) measurement technology (Genant et al., 1996). Due to the ability of DXA to accurately eliminate the soft-tissue absorption factor, BMD can be measured at central sites such as the whole body, lumbar spine and hip (see figure 1).



Figure 22. Picture of a hip scan from a DXA scanner.

There are both pencil and fan beam systems available. The fan beam is associated with a lower scan time, however the pencil beam has a lower dose of radiation. When using the pencil beam scanner, a child's hip scan takes approximately four minutes and has an effective dose of radiation of 1 μ Sv (unit for measuring ionising radiation effective dose which accounts for relative sensitivities of different tissues and organs exposed to radiation). The child's whole body scan takes up to 15 minutes and has an effective dose of radiation of 6 μ Sv. In comparative terms, this is less than natural background radiation received during the course of one day (up to 10 μ Sv) and much less than a chest or dental X-ray (up to 40 μ Sv, Ionising Radiation (Medical Exposure) Regulations, 2000).



Figure 23. Three participants holding pictorial representation of their hip scan, sitting on the DXA scanner.

Quality control procedures undertaken throughout all research consisted of system calibration at installation by a systems technician, to minimise variability among instruments, and daily quality control procedures. Daily quality control involved the scanning of an anthropomorphic phantom bone set in epoxy-resin, comparing scan results against known values and plotting data on a time line to enable the detection of trends or drifts in accuracy. The Hologic QDR system also uses an internal calibration system with a rotating filter of two sections of epoxy-resin-based material. At each measurement location the beam passes through the calibration system to provide constant calibration (Lohman, 1996).

As a measure of bone mineral content (BMC), DXA provides excellent accuracy, short-term precision and long-term reliability when these high quality control standards are met (Khan et al., 2001). Jergas and Genant (1993) reported the accuracy of the DXA to be between 90-99% (1-10% error around the actual value) and precision to be between 98-99% (1-2% error around repeated measurements). BMC as measured by DXA also correlates well with weight of cadaveric specimens; for example, a 94% of the variance in ash weight of infant swine piglets was explained by DXA measurements of whole body BMC (Hammami et al., 2001). The in-vivo precision error of DXA in our laboratory, expressed as the coefficient of variation, is approximately 1.0% for the total proximal femur and 0.5% for the whole body.

The limitations of DXA are inherent to all densitometers. DXA provides no measure of bone architecture. BMD is an areal density measurement (g.cm⁻²), i.e., BMC is divided by bone area, and therefore DXA provides a two-dimensional estimate of a three-dimensional structure. Consequently, throughout this thesis the research regressed BMC on bone area and body mass to produce a residualised measure of BMC, controlling for size differences between participants (Prentice et al., 1994). The last main limitation of DXA is that variability in results from intermanufacturer instruments can be as much as 20% (Genant et al., 1991). However, the research carried out in our laboratories all used the same instrument, the Hologic protocol for patient positioning standardised the analysis procedures using the same region of interest, and the same researcher conducted and analysed all scans.

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Appendix C: Assumptions for statistical tests



Appendix C: Assumptions of Statistical Tests

Based on Vincent (1999) and Stevens (1996).

Independent *t*-test

- The population from which the samples are drawn is normally distributed.
- The sample is randomly selected from the population.
- There is homogeneity of variance (the samples have approximately the same variance).
- The data is parametric.

Testing these assumptions

- Normality was tested by plotting a histogram with a normal curve to check for skewness.
- Homogeneity of variance was tested using Levene's test for equality of variance. If this assumption was violated (as in studies one, four and five) the degrees of freedom for the test statistic were reduced making the test more stringent.

Regression Analysis

- The ratio of subjects to independent variables should be no less than 5:1, and ideally approximately 20:1.
- The sample is representative of the population to which inferences will be made, and the relationship is linear within the population.
- The data is parametric.
- For each value of the independent variable the distribution of the dependant variable must be normal.

- There is homoscedasticity (the variance of the distribution of the dependent variable is approximately equal for all values of the independent variable(s)).
- There is no multicollinearity (the independent variables are not correlated). In the existence of multicollinearity there may be errors in signs and magnitudes of regression coefficient estimates. Consequently, in incorrect conclusions about relationships between independent and dependent variables may occur.

Testing these assumptions

- The ratio of subjects to independent variables was 57:3 or 19:1.
- Scatter plots of the independent variable against the dependant variable demonstrated a linear relationship.
- Normality was tested by plotting the residuals (the difference between the observed and predicted values of the dependent variable) on a histogram with a normal curve to check for skewness.
- A Q-Q plot of the standardised residuals was used to check if the sample came from a normal distribution (reflected by a straight line).
- A plot of the standardised residuals against the predicted values was used to check linearity and homoscedasticity (i.e. random scatter of residuals, if the residuals form a funnel shape this indicates heteroscedascity).
- Multicollinearity can be reflected in the standardised beta statistic and was
 examined using variation inflation factor (VIF). Variation inflation factor is the
 number of times the variance of the corresponding parameter estimate is
 increased due to multicollinearity. Values of VIF exceeding 2.5 may be a cause
 for concern regarding multicollinearity (Allison, 1999).

Analysis of Variance

- The sample is representative of the population to which inferences will be made.
- The dependent variable is parametric.
- The data is normally distributed.
- There is independence (there is no relationship between observations within and between groups).
- There is homogeneity of variance (the variance in the groups being tested are equal or nearly equal).

Additional assumptions for repeated measures and mixed model designs

• There is compound symmetry or sphericity (there is homogeneity of variance and homogeneity of covariance).

Testing these assumptions

- Normality was tested by plotting a histogram with a normal curve to check for skewness.
- Homogeneity of variance was tested using Levene's test for equality of variance. If this assumption was violated the degrees of freedom around the test statistic were reduced making the test more stringent.
- Compound symmetry or sphericity was tested using Mauchly's test. If this assumption was violated the Greenhouse Geisser (GG) correction factor was used to decrease the degrees of freedom around the test statistic, making the test more stringent.

Adapted Tukey's Test for Repeated Measures ANOVA

Significant interactions from the analyses of variance were followed up by the adapted Tukey's tests for repeated measures (Stevens, 1996, p.480);

 $|\chi_i - \chi_i| > q_{.05}; k, (N - J) (k - 1) \sqrt{(MS_{kxs/J}/N)}$

Where;

 $|\chi_i - \chi_j|$ is the magnitude of difference between the two groups, k is the number of within levels, N is the total number of subjects, J is the number of groups and MS_{kxs/J} is the error term.

For example (from study 3, calculating the critical difference between activity and monitor interactions for the X axis);

k is the number of activities = 6, N is the number of time samples = 10, J is the number of monitors = 8, $MS_{kxs/J}$ is the error term = 23752.468.

Degrees of freedom = 6, (10 - 8) (6 - 1)

6, 10 = 6.43 (read from the studentized range (q) (p = .01) (Vincent 1999, p.254) $\sqrt{(237522.468/10)}$

6.43 x 48.736 = 313.375

Tukey's critical difference is 313.4.

Therefore, when comparing activity counts recorded by monitors 1 and 2 at 4 km.h⁻¹, the difference between 765.2 and 750.1 does not exceed the critical difference of 313.4 and so they are not significantly different.

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Appendix D: Study 5

- 1) Institute of Health studies funding
- 2) Ethics approval
- 3) Informed consent forms
- 4) Parental information pack
- 5) Parental feedback

Athrofa Astudiaethau Iechyd The Institute of Health Studies

Gwenfro Building, Wrexham Technology Park, Wrexham, LL13 7YP. Tel: 01978 316238 (Sec.) Fax: 01978 311419 E-mail: HullLM@cf.ac.uk

15th November 2001

Dr Ann Rowlands School of Sport, Health and Exercise Sciences University of Wales Bangor George Building BANGOR LLS7 2PX

Dear Dr Rowlands

Re: Your Research Project: The relationship between physical activity, calcium intake, body mass and bone density in children

I am very pleased to be able to inform you that your submission for a grant from the North Walcs Health Research Trust Fund of £4,894.00 to support your research has been approved in full.

Would you please note the following requirements in relation to your grant:-

1) Would you please send all invoices to me for certification. I will then pass them on to the Treasurer's Department for payment.

2) Progress reports will be expected for each North Wales Research Committee meeting. These occur bi-annually and usually in March and October. I will inform you of the need for a report one month in advance of a meeting so that a progress report can be prepared for inclusion in the Committee's agenda papers.

3) A final report needs to be provided on completion of the research.

4) Acknowledgement of the support given by the North Wales Research Committee is expected in any publication of your research findings and a copy of the publication(s) should be provided to the institute when this is available.

Should you require any further help or should problems arise then please contact me. Otherwise may I wish you all the best with your project.

Yours sincerely

Dr. J.H.P. Evans Executive Secretary North Wales Research Committee

Cyfarwyddwr Astudiaethau: Director of Studies: Dr. J.H.P. Evans. Tel: 01978 316235 E-mail: JeffEvans@ihs.demon.co.uk

RESEARCH ETHICS COMMITTEE (WEST)

PWYLLGOR MOESEG YMCHWIL (GORLLEWINOL) AWDURDOD IECHYD GOGLEDD CYMRU

Ffon/Tel: (01248) 384877 (direct line)

Ffacs/Fax: (01248) 370629

Room 1/178 Ysbyty Gwynedd Bangor Gwynedd LL57 2PW

26th March 1999

Dr R Eston SHAPES University of Wales, Bangor Ffiiddoedd Building Victoria Drive Bangor Gwynedd LL57 2EN

Dear Dr Eston

Re : The effects of daily physical activity levels on bone mineral density and body fat in young children

Thank you for attending last night's Ethics Committee meeting to discuss your proposal.

The Committee considered this to be a most interesting and impressive application. As already discussed with you, it is imperative that studies of this nature involving children are conducted in line with stringent regulations. In order to safeguard potential participants, as well as yourselves as researchers, the Committee advises you arrange for all staff who will come into contact with the children during the course of the study to be subject to a screening process. Mr Bruce Napier has agreed to liaise with you to facilitate this.

Subject to this issue being addressed satisfactorily, receipt of a Welsh translation of the parent letter, and ratification on 15th April, the Committee is pleased to approve the conduct of this study.

I look forward to hearing from you

Yours sincerely

PP Dr DR Prichard

Chairman, Ethics Committee (west)

cc Dr A Rowlands

Member Organisations: Conwy & Denbighshire NHS Trust; North East Wales Institute of Higher Education; North East Wales NHS Trust;

Ysgol Gwyddorau Chwaraeon, Iechyd ac Ymarfer . Prifysgol Cymru, Bangor

Adeilad y George Bangor, Gwynedd LL57 2PX

Pfilm: (01248) 382756/383491 Swyddfa Gyffredinol Pfine: (01248) 371053 e-bost: shos@bangor.sc.uk http://www.shos.bangor.sc.uk



School of Sport, Health and Exercise Sciences University of Wales, Bangor

George Building Bangor, Gwynedd LL57 2PX

Tol: (01248) 382756/383491 General Office Fax: (01248) 371053 e-mail: shra@tungor.ac.uk http://www.shos.bungor.ac.uk

Dear Parent/Guardian,

I am a PhD student in the School of Sport, Health and Exercise Sciences at the University of Wales, Bangor. My research concerns the relationship between physical activity and bone mineral density in children. This is important as enhancing bone mass when young may help prevent the occurrence of osteoporosis and related fractures in later years.

We are looking for 60 girls and 60 boys, aged seven to nine years. The children taking part in this study will be required to wear a small activity monitor (the size of a pager, which clips on to a belt or waistband), for seven days. With parental help, all children will be required to keep a diary of all food and drink ingested over a four-day period and to fill out a maturational status form. At the end of the week the child's height, weight and bone mineral density (BMD) will be measured at the University. BMD will be measured using dual energy X-ray absorptiometry (DXA). This technique requires the child to lie still on a table while their body and hip are scanned. This will take approximately 10-14 minutes for the body scan and 5-5 minutes for the hip scan. Both procedures are completely pain free. Assessment of bone density by DXA is associated with exposure to X-rays, which is very small compared to other X-ray procedures, such as dental x-rays. It is also lower than the amount we are exposed to from background radiation in one normal day. From our experience, children find participating in these studies both rewarding and highly interesting. Information on your child's physical activity and BMD will be made available to you. Results may be used for publication with complete anonymity assured. Transport to and from the University will be provided.

Ethics approval has been granted by the North Wales Health Authority Research Ethics Committee (West) and this research is funded by the Institute of Health Studies.

If your child would like to participate in this study and you give your consent please complet the form below and return it to school. Your child's participation in this study would be greatly appreciated.

Yours faithfully,

Sarah Powell.

I certify that my child is participating in this study of the	eir own free will and that my child or I
may discontinue participation at any time. I give my co	nsent for my child to receive a DXA
body and hip scan at the University of Wales, Bangor.	
Name:	
Name of child:	Date of Birth:
Address:	i
Telephone number:	
	:
Signature of Parent/Guardian:	
Date:	
Please return to school as soon as possible. Thank you.	
•	

Ysgol Gwyddorau Chwaraeon. Iechyd ac Ymarfer Prifysgol Cymru, Bangor

Adeilad v George Bangor, Gwynedd LL57 2PX

Ffbs: (01248) 382756/383491 Swyddfa Gyffredinol Finer: (01248) 371053 a-boat shes@bangor.ac.uk http://www.shea.banaor.ac.uk



School of Sport, Health and Exercise Sciences University of Wales, Bangor

> George Building Bangor, Gwynedd LL57 2PX

Tel: (01248) 382756/383491 General Office Pax: (01248) 371053 nil: shos@hangor.ac.nir www.shes.bangor.ac.nk

Annwyl Riant/Gwarcheidwad.

Rwyf yn fyfyriwr PhD yn yr Ysgol Gwyddorau Chwaraeon, lechyd ac Ymarfer ym Mhrifysgol Cymru, Bangor. Mae fy ymchwil yn ymwneud â'r berthynas rhwng gweithgaredd corfforol a dwysedd mwynol esgym mewn plant. Mae hyn yn bwysig oherwydd gall cynyddu màs esgym yn ifanc fod o gymorth i rwystro osteoporosis, a thorri esgym sy'n gysylltiedig a hynny, rhag digwydd vn ddiweddarach mewn bywyd.

Rydym yn chwilio am 60 o enethod a 60 o fechgyn saith a naw oed. Bydd angen i'r plant a fydd yn cymryd rhan yn yr astudiaeth hon wisgo monitor gweithgaredd bychan (o faint pager, sy'n clipio ar fand gwasg neu wregys) am saith diwrnod. Gyda chymorth rhieni, bydd angen i bob plentyn gadw dyddiadur o bob bwyd a diod gaiff ei fwyta/yfed dros gyfnod o bedwar diwrnod a llenwi ffurflen statws seddfedol. Ar ddiwedd yr wythnos mesurir taldra, pwysau a dwysedd mwynol esgym (BMD) y plentyn yn y Brifysgol. Mesurir BMD trwy ddefnyddio pelydr-X absorptiometreg (DXA) ynni deuol. I wneud hyn mae angen i'r plentyn orwedd yn llonydd ar fwrdd tra caiff y corff a'r glun eu sganio. Bydd yn cymryd tua 10-14 munud i sganio'r corff a 5-6 munud i sganio'r glun. Nid yw hyn yn boenus o gwbl. Wrth asesu dwysedd esgym 1 DXA mae'r pelydr-X yn fychan iawn o'i gymharu & dulliau pelydr-X eraill, megis pelydr-X deintyddol. Mae'r ymbelydriad yn is hefyd na'r hyn a gawn trwy ymbelydriad cefndirol mewn diwrnod arferol.

O'n profiad ni, mae plant yn cael budd wrth gymryd rhan yn yr astudiaethau hyn ac yn meddwl eu bod yn ddiddorol iawn. Bydd gwybodaeth am weithgaredd corfforol a BMD eich plentyn ar gael i

chi. Gall canlyniadau gael eu cyhoeddi ond ni ddatgelir unrhyw wybodaeth am y rhai a gymerodd ran. Darperir cludiant i'r Brifysgol ac yn ôl.

Mae'r ymchwil hon wedi cael ei chymeradwyo gan Bwyllgor Moeseg Ymchwil Awdurdod lechy Gogledd Cymru (Gorllewin) ac fe'i cyllidir gan y Sefydliad Astudiaethau Iechyd.

Os hoffai eich plentyn gymryd rhan yn yr astudiaeth hon a'ch bod chi'n rhoi eich caniatåd a fyddech cystal & llenwi'r ffurflen isod a'i dychwelyd i'r ysgol. Gwerthfawrogir cyfranogiad eich plentyn yn yr astudiaeth hon yn fawr iawn.

Yn gywir

Sarah Powell

Rwy'n tystio bod fy mhlentyn yn cymryd rhan yn yr astudiaeth hon o'i (g)wirfodd ac y gall ef/hi roi'r gorau i gymryd rhan unrhyw bryd. Rwyf yn rhoi caniatâd i'm plentyn gael sgan corff a chlu ym Mhrifysgol Cymru, Bangor,

Enw:

Enw'r plentyn:

Cyfeiriad:

Rhif Ffôn

Llofnod Rhiant/Gwarcheidwad

Dyddiad:

Dychwelwch i'r ysgol cyn gynted â phosibl os gwelwch yn dda. Diolch.

Dyddiad Geni:

Ysgol Gwyddorau Chwaraeon. Iechyd ac Ymarfer Prifysgol Cymru, Bangor

Adeilad y George Bangor, Gwynedd LL57 2PX

Ffba: (01248) 382756/383491 Swyddfa Gyffrediaol Fface: (01248) 371053 e-bost: shes@bangor.ac.uk http://www.shes.bengor.ac.uk

UNIVERSITY OF WALES BAN GOR

• PRIFYSCOL CYMRU •

School of Sport. Health and **Exercise Sciences** University of Wales, Bangor

George Building Bangor, Gwynedd LL57 2PX

Tel: (01248) 382756/383491 General Office Fax: (01248) 371053 il: shes@baseor.ac.uk http://www.ahee.he meor.ac.uk

20th May 2002

Dear Parent/Guardian

Thank you for your interest in the bone density research we are carrying out at the School of Sport. Health and Exercise Sciences. It has come to our attention that a parent has expressed some concern regarding scanning the children on the dual energy x-ray absorptiometer (DXA). We hope this letter will reassure you about the safety of our procedures and the value of this important research.

It is important to note, from the outset, that all of our projects involving children and DXA methods have been fully approved by the North West Wales Area NHS Ethics Committee for human research. This committee is comprised of medical experts and is fully aware of the nature of DXA scanning. The Ethics Committee would clearly not approve any research if it doubted first and foremost, the safety of the incasurement methods, and secondly benefits and viability of the research project. Additionally, this particular project is funded by the North Wales Research Committee, a Sub Committee of the North Wales Health and Social Care Research and Development Collaboration.

The machine we use for DXA scanning is a pencil beam scanner. We have consulted widely on the effective dose received by a child using this technology. This has included reference to the Ionising Radiation (Medical Exposure) Regulations (IRMER, 2000) and professional consultation with Dr Glen Blake at the Department of Nuclear Medicine at Guy's Hospital, London. The IRMER 2000 details indicate that for a spine and femur (hip) DXA the pencil beam delivers an effective dose of 1 µSV, and for the total body 6 µSv. According to the IRMER guidelines, one day's normal background radiation is equivalent to 10 uSy. There is therefore an extremely negligible risk. It is less than standing outside for a day.

We also confirm that all staff eligible to use the DXA are appropriately certified and trained to use the equipment. They have received a training course from Vertee (the company supplying and maintaining the machine), and have attended both POPUMET (Protection of Persons Undergoing Medical Examination or Treatment) and the more recent 2000 IRMER (Ionizing Radiation (Medical Exposure) Regulations) course. We also confirm that we adhere to the administrative, staffing, bureaucratic, technical, service and all health and safety requirements surrounding the use of the DXA in SHES.

Nevertheless, health and safety aspects surrounding its use, and all other equipment and procedures applied to human research, are continually reassessed to ensure the maximum protection for both tester and volunteer participant.

We hope this information is useful to you and satisfies you as to the safety of our procedures. Sarah Powell will contact you shortly to discuss participation in this project. We would be pleased to provide further detail on the above procedures surrounding the use of the DXA or any other details regarding this project.

Yours sincerely.

₹

Professor Roger Eston (Head of School)

Dr. Ann Rowlands (Project Supervisor, 01248 383486) Miss Sarah Powell (Researcher, 01248 388147)

The relationship between physical activity, calcium intake, body mass and bone density in children.

Lifetime risk of osteoporosis and related fractures depends on the peak bone mass achieved at skeletal maturity, and subsequent age-related bone loss (Bachrach, 2001¹). There is no cure for osteoporosis once the disease is established, and a large amount of bone will have already been lost by the time of fracture (Lysen and Walker, 1997²). At least 90% of total bone mass is accrued by the end of adolescence (Glastre et al., 1990³). Therefore, it is important to optimise early bone accrual in order to reduce subsequent loss.

As a result we hope to determine the relationship between physical activity, calcium intake, and bone mass in pro-pubertal children. We aim to identify the quantities of physical activity and calcium necessary for optimisation of bone mass in childhood.

Please find overleaf one day's physical activity assessment and detail of the DXA hip and whole body scan.

³ Glastre C, Braillon P, David L, Cochst P, Meunier PJ, Deimas PD 1990 Measurement of bone mineral content of the humber spins by dual energy x-ray absorptiometry in normal children: correlation's with growth parameters. J Endocrinol Metab 70:1330-1333.



¹ Bachrach LK 2001 Acquisition of optimal bone mass in childhood and adolescence. Trends Endocrinol Metab 12:22-28. ² Lysen VC, Walker R 1997 Osteoporosis risk factors in eighth grade students. J Sch Health 67:317-322.



This is a typical daily physical activity routine for a 9 yr old boy. As you can see his day starts at 8.30 and details activity spurts, which coincide with his journey to school, school breaks and after school activity- his day then finished at 21.30. This data will be analysed for total activity (average counts per day) and minutes spent in vigorous, moderate and low intensity activity.

DXA scan

Each child will be brought to the University with a parent or a friend from school to have two scans. The hip scan takes approximately 5 minutes and the whole body scan takes approximately 15

minutes. Please see pictures of the two scans below.



The effective dose of radiation for the hip scan is no more than 1 μ Sv (microSievert) and the whole body scan no more than 6 μ Sv. This totals 7 μ Sv which is equivalent to less than 1 day's normal background radiation (see below). It is much lower than a chest x-ray. The extracts below are from the IRMER course (Ionising Radiation Medical Exposure Regulations 2000).



Some activities carrying a risk of death comparable to receiving an effective dose of 10 µSv from a DXA scan

- Exposure to natural background radiation for 2
- days
- Smoking a cigarette
- •Travelling 30 miles by car
- •Travelling 150 miles by acroplane
- •Working in a factory for a week
- •Being a woman aged 50 for 1 hour



Ionising Radiation (Medical Exposure) Regulations 2000: a training course for practicioners and operators involved in bone densitometry Ysgol Gwyddorau Chwaraeon Iechyd ac Ymarfer Prifysgol Cymru, Bangor

Adeilad y George Bangor, Gwyneidd IL57 2PX Ffön: (01248) 382756/383491 Swyddfa Gyffredinol Ffacs: (01248) 371053 E-bost: shes@bangor.ac.uk http://www.shes.bangor.ac.uk School of Sport, Health and Exercise Sciences University of Wales, Bangor

George Building Bangor, Gwynedd LL57 2PX Tel: (01248) 382756/383491 General Office Fax: (01248) 371053 E-mail: sbes@bangor.ac.uk http://www.shes.bangor.ac.uk



Dear

I would firstly like to thank you for your participation in this study it is much appreciated. A total of 110 children took part in this study during the months of March through to September. The average age was 9 years (mean \pm SD, 9.03 \pm 0.869), height was 132.24 cm (132.24 \pm 6.66) and weight was 31.18 kg (\pm 1.18 \pm 7.14). Please find enclosed, physical activity graphs, drawn from data downloaded from the monitor. Below is a list of the

days your child wore the monitor, detailing how long they spent in varying intensities of physical activity:

Day	Low	Moderate	High 📜	Vigorous	[[
•	Intensity	Intensity	Intensity	Intensity	ŀ
1					
2					
3	•				
4					
5					
6					
7		·			

To meet the National Health Education Authority's guidelines for physical activity:

"A child must participate in at least 60 minutes of moderate or above intensity activity per day". (Health Education Authority. Young and Active? Young People and Health Enhancing Physical Activity- Evidence and Implications. London: Health Education Authority, 1998, pp.171). Please find an example of each intensity below: Low; playing catch, Moderate; walking at 4km per hour, High; walking at 6km per hour,

Vigorous; running at 8km per hour or playing hopscotch.

A greater bone mass gained early in life is now considered a critical factor in protecting against osteoporotic fractures later in life. The critical years for skeletal growth and accumulation of bone mass lie in the preputertal and pubertal decades (Anderson, J.J.B. 2000. The important role of physical activity in skeletal development: how exercise may counter low calcium intake. American Journal of Nutrition: 71:1384-1386). Please find enclosed a picture of the full body scan, detailing total bone area (BA), bone mineral content (BMC) and bone mineral density (BMD). The mean bone mineral density in this sample of children was 0.839 ± 0.052 g/cm². The BMD will differ between children of a similar age as BMD depends highly on height, weight, pubertal stage, activity level and nutrition,

which will be strictly controlled for during the statistical analysis of the data.

Please note that unless all food ingested were weighed and reported the daily calcium content can only be an estimation. If the weight of all food ingested was reported and the calcium content is lower than recommended, it can be raised by the ingestion of foods such as; yogurt, milk, cheese and ice cream!

I hope this information was of interest to you, once the data has been analysed and conclusions have been drawn a summary will be sent to you and a poster will be presented to the school.

Thanks again for all your help.

Sarah Powell.