

Material and methods: 38 male patients fulfilling the New York criteria of AS aged 23-67 years with disease duration of 3-30 years, underwent QCT (lumbar spine) and DXA (lumbar spine and proximal femur) measurements and x-ray (thoracic and lumbar spine). 12 male patients with lumbar spine osteoarthritis served as controls for QCT evaluation.

Results: Lumbar spine trabecular BMD (region of interest) measured by QCT correlated significantly only with BMDs of Ward's triangle measured by DXA ($r=0.63$; $p<0.05$). Both measurements (lumbar spine QCT and Ward's triangle DXA) correlated with disease duration ($r=-0.51$; $p<0.05$, and $r=-0.37$; $p<0.05$ respectively), however only QCT values of the lumbar vertebra negatively correlated with the radiological grading of disease progression at spine.

Conclusions: Our data indicate that in AS only trabecular bone BMD assessment regardless of the technique, i.e. DXA or QCT, carries valuable information which correlates with the duration of disease and radiological grading of the spine. The DXA assessment of lumbar spine or femoral neck possess a significant accuracy error due to perivertebral calcification of the soft tissues and has no value for monitoring BMD changes in AS.

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PERFORMANCE AND LIMITS OF IN VIVO MICRO-CT IMAGING OF TRABECULAR BONE IN RATS AND MICE, WITH CONSIDERATION OF ANIMAL WELFARE AND TISSUE-WEIGHTED DOSIMETRY

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Important advantages to experimental design of preclinical bone studies are available, if trabecular bone of rats and mice can be imaged *in vivo* under anaesthesia. However, *in vivo* scanning of rodents imposes constraints on micro-CT scanner design, measurement geometry and imaging parameters, with the result that the 3D x-ray image resolution is reduced compared to that attainable in *ex vivo* scanners. The specific constraints include (a) rotating source-detector assembly around immobile subject, (b) fixed source-detector separation, (c) loss of image contrast due to x-ray absorption in live soft tissue surrounding bone, (d) limits to radiation dose to the animal, and (e) breathing and heart-related movement of the animal during scanning. A further constraint is stress to animals and consequent weight loss caused by anaesthesia, an important consideration when contemplating multiple sequential scans. We demonstrate that adequate resolution of trabecular bone structures can be achieved in rodents *in vivo*, while not compromising animal welfare by excessive periods of anaesthesia or excessive radiation dose. The scanner employed was the Skyscan 1076. 3D images with pixel size of 8.9 microns (Gaussian resolution of 12.5 microns) can be obtained at the rodent hindlimb knee with local absorbed dose of about 0.6 Gray and whole body effective dose equivalent of 10 milliSieverts (using ICRP tissue weighting factors). This resolution allows structural histomorphometric analysis of rat and mouse trabecular bone. X-ray source shuttering can reduce dose by a further 50%. The sensitivity of micro-CT-measured bone parameters to changes in resolution and contrast are systematically evaluated. Parameters vary in sensitivity, with connectivity parameters such as Euler connectivity being especially sensitive to resolution. It emerges that the fixed scanning parameters of the *in vivo* scanner are partly advantageous in that they impose standardisation on scanned images and calculated structural parameters of trabecular bone.

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MICROMECHANICS OF OSTEOPOROTIC BONE MEASURED BY SCANNING ACOUSTIC MICROSCOPY

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The Scanning Acoustic Microscopy (SAM) reveals the possibility to deliver information on elastomechanical properties. The sound velocity depends on elastic moduli and the elastic stiffness directly correlates with the sound velocity in the direction of propagation. We used the acoustic impedance as a suitable parameter for characterization of bone. Latest SAM-devices with frequencies up to 2 GHz gain a spatial resolution of about 0.5 micrometer, comparable with the light microscopy, which allows distinction of cellular and subcellular structures. We developed the image processing software Multi Layer Analysis (MLA) to exclude the influence of surface topography and to focus the images for quantitative investigation at frequencies up to 900 MHz. Consecutively a well focussed output image was calculated by MLA finding the maximum $V(z)$ -position for every pixel. The mechanical properties of osteoporotic bone were measured in iliac crest biopsies. The bone was embedded in PMMA and specimens with planparallel surfaces had been prepared by grinding and polishing. The acoustic procedure is nondestructively, the correlation with nanoindentation measurement of elastic moduli at the same location allows the calculation of material parameters. The results of impedance measurement in osteoporotic and normal bone will be presented at the session. Besides measurement the images deliver morphological information. Especially the transition of regions containing hydroxyapatite minerals to neighbouring structures of organic components and the process of maturation become clearly visible, contrasted without any staining or alteration of the tissue. By means of image processing we gain well

focussed images and an extended procedure of calibrating ensures the reproducibility of measurements. Contemporary trials to model bone in biomechanical theories and to judge its loading capacity reveal a lack of measuring procedures, capable to deliver information on bulk properties of bone and on the distribution of these properties in two or three dimensions. But even loading capacity of bone becomes more and more interesting to evaluate the quality of bone in case of osteopathies. Scanning acoustic microscopy seems to be a valuable tool to detect elastomechanical properties via measurement of acoustic parameters.

Osteoporosis: Pathophysiology, Genetics, Epidemiology

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BONE DENSITY AND MENOPAUSE AGE IN WOMEN WITH A HISTORY OF ACNE VULGARIS: A PRELIMINARY STUDY

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Adequate bone mineral density (BMD) and oestrogen levels are linked. Increased androgen levels are associated with decreased bone loss in women, and therapeutic androgens have been used in osteoporosis prevention. High levels of androgens also occur in women with severe acne vulgaris. It was hypothesised that women with a history of acne vulgaris might have better BMD than women without this history.

Method: Questions concerning a history of acne vulgaris were included in a health questionnaire normally administered to women referred to a bone densitometry unit in the NE of England. Women with a history of acne rosacea were excluded from the study. 584 post-menopausal women were included in this study. Data included BMD (gm/cm^2) of lumbar spine and hip (DEXA), age, weight, height, age at menopause, history of osteoporosis and history of HRT.

Results and discussion: 12.8% (75/584) women reported a history of acne vulgaris, 20% (15/75) of whom had a history of HRT. Of the 'acne free' group 18.4% (94/509) had HRT. No differences between the two groups emerged for age, weight or height. As expected for all subjects there was a significant relationship ($p<0.01$) between premature onset of menopause (age <45 y) and osteoporosis. However, significantly fewer women with acne history ($p<0.01$) had premature onset compared with those who were 'acne free' (29.7% against 43.3%); and of the 22/75 (29.7%) 'acne history' women with premature onset, 64% (14/22) had surgically induced menopause. Overall, 20.3% of the women with acne history had late onset menopause (>52 y) compared with only 9% of those who were 'acne free'. The BMD of lumbar spine and hip were significantly greater in women with a history of acne ($p<0.04$). The level of significance increased ($p<0.02$) when women with an HRT history were excluded from the analyses. Women with acne also had a lower incidence of diagnosed osteoporosis than the 'acne free' group (8% against 14%). In conclusion, women with acne history in this study had later onset of normal menopause and better BMD than those who were 'acne free'. These results however are derived from a limited sample. This apparent link therefore requires more extensive investigation.

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HYSTERECTOMY, BMI AND BMD IN LEBANESE POPULATION

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Bone mineral density [BMD] increases during growth until a peak is reached at maturity. The risk of development of osteoporosis post surgical menopause [hysterectomy] depends on the peak bone density and the rate of its subsequent bone loss. To identify whether a high Body Mass Index [BMI] could affect the BMD, we compared body size and BMD levels among 320 women underwent hysterectomy for various length of time [up to 39 years]. There was a direct correlation between BMI and BMD in all groups at any age and for any length of time. Women with high BMI who underwent hysterectomy [without oophorectomy] maintained a higher BMD levels than those with low BMI.

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INFLUENCES OF THE VITAMIN D AND ESTROGEN RECEPTOR GENES ON BONE MINERAL DENSITY IN POSTMENOPAUSAL WOMEN IN MALTA

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Polymorphisms in the vitamin D (VDR) and estrogen receptor (ER) genes have been associated with bone mineral density (BMD) and an increased risk of osteoporosis. In this study, a start codon polymorphism (FokI) and polymorphisms at the 3' end (BsmI, ApaI, TaqI) of the VDR gene, together with two polymorphisms in intron 1 of the ER gene (PvuII, XbaI) and their interactions were analysed in postmenopausal women in Malta.

108 postmenopausal Maltese women (55.1 ± 6.9 years) were recruited for this study. Polymorphisms in the VDR and ER genes were analysed by PCR restriction fragment length polymorphism (RFLP) while BMD at the lumbar spine and femur was measured by DEXA.

Allele frequencies for VDR RFLPs observed were as follows: F 75.7%, f 24.3%, B 42.3%, b 57.7%, A 58.4%, a 41.6%, T 58.6%, t 41.4%; and for ER RFLPs were: P 42.1%, p 57.9%, X 37.5%, x 62.5%, and all were in Hardy-Weinberg equilibrium. Polymorphisms at the 3' end of the VDR gene were found to be in strong linkage disequilibrium with each other when tested by chi-squared test ($p < 0.001$) but were in equilibrium with the start codon polymorphism ($p > 0.05$). Intron 1 polymorphisms of the ER gene were also in strong linkage disequilibrium ($p < 0.001$) with each other. The highest BMD at both anatomical sites was observed in FF homozygotes and PP homozygotes although no statistical significance was reached (ANOVA FokI: Lumbar $p = 0.375$, Femoral $p = 0.405$; PvuII: Lumbar $p = 0.769$, Femoral $p = 0.803$) even after adjustment for age, BMI and years since menopause. The most frequent genotypes for the VDR gene were BbTtAa 33.3%, BbttAA 17.6%, bbTTaa 16.7%, BbTtAA 14.7%; while for the ER were PpXx 41.7% and ppxx 32.4%. These genotypes were distributed similarly in normal, osteopenic and osteoporotic women (Chi-squared: VDR gene: $p = 0.933$; ER gene: $p = 0.932$). No statistically significant difference was reached for BMD between individuals of different genotypes even after combining genotypes within the same gene and between the two genes.

In conclusion, VDR and ER genes do not seem to have any effects on BMD in postmenopausal women in Malta.

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QUANTITATIVE ULTRASOUND OF THE OS CALCIS OF SUB-SAHARAN AFRICAN MEN: AGE-RELATED CHANGES, PREVALENCE OF OSTEOPENIA AND OSTEOPOROSIS

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Fragility fractures are reported to be rare among both men and women in sub-Saharan Africa. Reasons to this apparent low incidence of fractures among Africans remain unclear. Limited data available suggests a normal or lower bone mineral density among Africans women compared to other populations. Presently, there are no studies looking at sub-Saharan African men. This community-based cross-sectional study, using a Lunar Achilles Express Ultrasonometer, assessed age-related changes of heel QUS parameters and the prevalence of osteopenia and osteoporosis among a convenience sample 220 men, ages 30 to 91, in Cameroon, Central Africa. Men aged 30-39 ($n=58$) were selected to form the QUS peak bone mass group (PBM). Interestingly, this PBM and derived t-score did not differ from the machine reference values established from North American Caucasian women. In the total study group, the prevalence of osteoporosis and osteopenia using WHO criteria was nearly the same using the machine reference or our calculated t-scores of respectively 5% and 22.3%. The prevalence of osteoporosis rises from 0% in the 30-39 year old group to 40% after age 80 indicating a decline in bone quality comparable to what occurs in Western countries. However, the age-dependent decline pattern of stiffness was quite different; showing two distinct phases. A relatively stable plateau from the 4th to the 8th decade with only 10.9% decrease in the SI; followed by an exponential loss in bone quality occurring later, with 42.9% decrease in the SI between the 8th and the 10th decade. While the Stiffness Index-peak does not appear to be increased in sub-Saharan African men compared to western countries, a more stable bone density and quality as reflected by this QUS parameter, till age 80 may play an important role in the lower incidence of fractures seen in this population. Further investigation with longitudinal monitoring should be performed to investigate this hypothesis resulting from a cross-sectional observation.

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THE EFFECT OF AGE ON SERUM LEVELS OF OSTEOPROTEGERIN AND RANKL

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Background: RANKL (receptor activator of nuclear factor κ B ligand) and osteoprotegerin are central regulators of osteoclast generation. Since elderly subjects are known to be at risk for osteoporotic fractures, we were interested in the effect of age on levels of osteoprotegerin and RANKL.

Subjects and methods: In this study we included 25 young women (mean age: 30 ± 4 years), 25 young men (mean age: 31 ± 5 years), 28 elderly women (mean age: 85 ± 7 years) and 34 elderly men (mean age: 82 ± 12 years). Serum levels of osteoprotegerin, RANKL, sex hormones and markers of bone metabolism were measured by ELISA or RIA.

Results: We found significantly decreased levels of 25OH-vitamin D3 and increased levels of parathyroid hormone in the elderly when compared to the young subjects. Moreover, levels of osteocalcin and c-terminal telopeptide of type I collagen (CTX) were higher in aged men and women than in young controls. Serum levels of osteoprotegerin were significantly higher in the elderly than in the young subjects. In contrast, serum levels of RANKL did not change with age. As expected, levels of sex hormones significantly decreased with age in elderly men and women. Interestingly, elderly men had higher 17-beta-estradiol levels than elderly women.

Conclusion: Increased bone remodelling in elderly subjects appears to be due to sex hormone deficiency and secondary hyperparathyroidism. Serum levels of osteoprotegerin, but not those of RANKL increase with age in men and women.

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THE EFFECT OF DIET ON BONE MINERAL DENSITY AND SERUM INSULIN-LIKE GROWTH FACTOR-1 LEVELS IN PERI- AND POSTMENOPAUSAL WOMEN

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Background: Osteoporosis is a metabolic bone disease that affect commonly postmenopausal women and elderly population.

Aim: To evaluate the relationships between nutritional intake and bone mineral density (BMD) and determine the serum insulin-like growth factor-1 (IGF-1) levels in peri- and postmenopausal women.

Methods: 44 women (25 premenopausal, 19 postmenopausal) with ages between 40-60 years were enrolled. Daily mean intakes of different nutrients were obtained with seven-day dietary record. BMD's were acquired by DXA, and serum IGF-1 levels with RIA. Associations between mean daily nutritional intakes and lumbar and femoral BMD's and serum IGF-1 levels were investigated.

Results: No relationship was found between daily protein and other dietary intakes and lumbar and femoral BMD's. Serum IGF-1 levels were found to be independent from daily protein intake and showed no association with lumbar and femoral BMD's.

Conclusion: It's seemed that, nutritional factors in a normal mixed diet does not affect mainly the manifestation of osteoporosis in women. There is need for more studies with larger series to evaluate the role of IGF-1 system in this relationship.

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THE INCIDENCE AND DIAGNOSTIC CRITERIA OF PRIMARY OSTEOPOROSIS IN CHINA

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From over 10 years clinical practice and epidemiological studies, we realize that the appropriate diagnostic criteria are the most important thing among basic research, clinical studies of the prevention and therapy for osteoporosis. In this paper, we will discuss the following two problems for using dual energy X ray absorptiometry (DEXA).

Which part of the bone is the better place to be measured? We can easily find that the results of bone mineral density in different areas are different. One of the reasons is the effect of bone size, shape and the direction. Hip neck is considered to be the best place for BMD measurement and osteoporosis diagnosis, because it has less artificial influences from the analysis based on our data. And we can see that ward's area (early bone loss, $>25\%$ at age 50) can not provide the accurate data for osteoporosis diagnosis. For L2-4 one should consider that soft tissue calcification can cause artificial high reading.

In 1985, the WHO proposed the following osteoporosis diagnostic criteria: a T-score of minus 2.0 SD below the mean BMD value for a young adult of the same sex was characterized as osteoporosis. In 1994, Dr. John A. Kanis advanced the following osteoporosis diagnostic criteria: osteoporosis-a value for BMD or BMC more than 2.5 SD below the young adult average value. In Japan, Dr. Orimo and Japanese bone mineral society defined the osteoporosis diagnostic guidelines. If BMD is 70% below the mean value of young adult of the same gender, it is osteoporosis. We suggest the following criteria for Chinese people: the cumulated bone loss $>25\%$ than the mean value of young adult of the same gender in Chinese can be diagnosed having osteoporosis.

Based on the above-mentioned results, in China, women over age 60 years and men over age 75 years have osteoporosis. Based on the fifth population census, conducted in 2000, the population of women over 60 years old is 75.57 million; and that of men, over 75 years old is 12.69 million; the total osteoporosis is 88.26 million,