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# IMPACT OF MINERAL CONTENT IN HUMAN TRABECULAR BONE ON ITS ELASTIC PROPERTIES

## A. Mazurkiewicz<sup>\*</sup>

**Abstract:** The paper discusses the results of measurement of elastic modulus and the mineral content for 42 samples of trabecular bone, collected from femoral heads after alloplastic implantation of hip joint. The samples were collected from femoral heads with coxarthrosis and osteoporosis. Measurement of elastic modulus in compression test, and with the use of ultrasonograph technique with 0.5, 1 and 2 MHz frequency of transducers was performed for the samples. Also, evaluation of mineral density of the bone - BMD, and ash density - Ash.D was performed. The results obtained suggested usability of BMD and Ash.D indicators for assessment of elastic properties of trabecular bone. The relations with elastic modulus evaluated using R correlation coefficient were similar for BMD and Ash.D, and contained within the range R = 0.57 - 0.68.

Keywords: Trabecular bone, Elastic modulus, Compression test, Ash density, BMD.

## 1. Introduction

Trabecular bone is a part of the bone, the quality of which has high significance for its strength. It is a spacial structure of trabeculae built of colagen fibres and mineral crystals seated on them, mainly hydroxyapatite. The amount of minerals contained in the structure is important for its mechanical properties (Mosekilde et al., 1987 and Yuehuei and Draugh, 1999). Reduction in the amount of mineral components with age or during bone diseases leads to loss of bone mass, and, consequently, to loss of strength properties of the whole bones (Covin, 1999). "In vivo" bone quality assessment is performed mostly through densitometric examination. By comparing the result obtained with values adopted as standard for the given gender and age in the given population, it can be evaluated whether the bones have sufficient strength. One of the indicators obtained from the test is bone mass density - BMD. The measurement is performed through the skin and soft tissues, and so it contains certain inaccuracies resulting from the individual characteristics of patients. The actual number of mineral components contained in the bone can be evaluated "in vitro" based on ash density measurement - Ash.D. To do so, the sample must be incinerated in order to burn out the organic part of the bone, i.e. colagen fibres. After this, only the mineral components of the bone will remain. Comparison of mass before and after incineration will allow for quantitative evaluation of the mineral phase content in the sample. This type of examination may only be performed "in vitro". It is difficult to definitely determine the relationship between these indicators and the mechanical properties of bones, since the former is measured "in vivo", while the latter - "in vitro". Thus, the aim of this paper was to determine the usability of both these indicators for evaluating elastic properties of trabecular bone. The value of elastic modulus of trabecular bone was assumed to be the measure of elastic properties. The modulus was evaluated using two methods: in a compression test, and through the ultrasonographic method, with the use of transducers of three different frequencies. Ash density and BMD evaluations were performed on samples collected from human femoral heads. The BMD measurement was performed on a clinical densitometer, as "in vitro" examination of the samples. A protocol allowing for simulation of such examination performed "in vivo" was used for the measurement.

<sup>&</sup>lt;sup>\*</sup> Assoc. Prof. Adam Mazurkiewicz, PhD.: Department of Mechanical Engineering, University of Sciences and Technology, Kaliskiego 7 Street; 85-789 Bydgoszcz; PL, e-mail: adam.mazurkiewicz@utp.edu.pl

### 2. Methods

42 samples were collected from ostheoporotic and coxartric femoral bones obtained as a result of hip joint implanting. The age of the patients ranged from 51 to 82 years with an average of 70 years. The samples used for the tests were not divided according to the type of disease or the age of patients. The samples were stored in 10 % formalin solution at the room temperature (Edmondston et al., 1994). Samples collecting scheme is shown in Fig. 1.



Fig. 1: Samples collecting scheme.

BMD measurement was performed using Lunar Expert (General Electric) clinic-grade densitometer, according to the procedure specified by the manufacturer in the technical documentation of the device for bone mass density measurements on bone samples, performed "in vitro". The procedure includes simulating the presence of soft tissues surrounding the bone, same as in "in vivo" evaluation. BMD values were obtained in g/cm<sup>2</sup>.

Elastic modulus measurement on the sample was performed with the use of ultrasonographic method, and through a non-destructive compression test. To perform the USG measurement was used Panametrix 5058RP generator. The samples were placed in a vacuum pump for 30 minutes in order to remove air from pores lying near the sample surface. Then, the samples were put under water. Longitunal waves transducers of 0.5, 1 and 2 MHz frequency were used for the measurements. The sending and receiving transducers were placed near head surfaces of the sample. Wave passage time through the sample was measured, and - based on this - the value of elastic modulus of the trabecular bone was calculated. During the measurement, the transducers and the sample were kept in a water tank in order to prevent penetration of air to pores of the sample.

The compression test was performed using testing device Instron E3000 as follows: the sample was placed in a testing machine. Initial load was 3 N. Next five preliminary cycles were performed until reaching deformation value  $\varepsilon = 0.3$  % of sample height, in order to stabilize the surface of contact between head surfaces of the sample and working surfaces of the machine. The next cycle was performed until obtaining deformation equal  $\varepsilon = 0.8$  %, i.e. the limit of elastic deformation for trabecular bone (Yueuhei et al., 1999). After this, value of secant elastic modulus was calculated from deformation range  $\varepsilon = 0.2 - 0.8$  %.

In order to perform the ash density measurement, the samples were dried in a temperature of 120 °C for 24 hours, and were weighted afterwards. Then, they were incinerated in a furnace, in a temperature of 800 °C for 18 hours, according to the procedure described by Yuehuei et al. (1999). After this, the samples were weighted again. The mass of each sample after incinerating was divided by its initial volume - before incinerating. This way, Ash.D value in  $g/cm^3$  was obtained.

Additionally, mass content of mineral components in the dry sample, marked as %Min, was measured. The value was calculated as a quotient of sample mass before and after incineration (but after drying in 120 °C), and expressed in percents.

## 3. Results

Tab. 1 presents the results of measurements performed. The range of values obtained, mean result, standard and relative standard deviation is presented. Tab. 2 presents values of coefficients of correlation between BMD values, Ash.D, % Min, and the modulus values from compression tests – marked as Mmt and measurements with USG method at transducers frequency: 0.5 MHz -  $M_{0.5}$ , 1 MHz -  $M_1$  and 2 MHz -  $M_2$ .

	<b>Age</b> [year]	<b>BMD</b> $[g/cm^2]$	<b>Ash.D</b> $[g/cm^3]$	<b>%Min</b> [%]	M <sub>Mt</sub> [MPa]	<b>M</b> <sub>0.5</sub> [MPa]	M <sub>1</sub> [MPa]	M <sub>2</sub> [MPa]			
Min	51	0.136	0.114	15.95	41.3	1612.3	1646.7	1706.4			
Max	82	0.432	0.592	65.54	1306.4	7093.4	8601.4	11029.7			
Av	70.0	0.264	0.302	33.22	391.7	4594.1	5150.2	5960.6			
SD	10.4	0.078	0.087	7.93	263.3	1432.8	1551.3	1912.1			
RSD [%]	15.0	29.7	28.8	23.9	67.2	31.2	30.1	32.1			
where: Min – minimum, Max – maximum, Av – average, SD – standard deviation, RSD – relative standard deviation.											

Tab. 1: Values of density and elastic moduli obtained from the measurements.

Tab. 2: Values of the obtained coefficients of correlation R
between different densities and elastic moduli.

	M <sub>Mt</sub>	$M_{0.5}$	$\mathbf{M}_{1}$	$M_2$
	[MPa]	[MPa]	[MPa]	[MPa]
<b>BMD</b> $[g/cm^2]$	0.57	0.64	0.62	0.64
<b>Ash.D</b> $[g/cm^3]$	0.66	0.68	0.61	0.63
<b>%Min</b> [%]	0.47	0.32	0.30	0.31

Fig. 2 present example of relationship between ash density and  $M_{0.5}$  modulus measured through USG method at 0.5 MHz frequency.



*Fig. 2: Relation between ash density and*  $M_{0.5}$  *module measured by USG method.* 

#### 4. Conclusions

The values of compression moduli from the compression test and USG method correlate with the values obtained by other authors for the given test method (Goldstein, 1987, Ashmann et al., 1988, Rho et al., 1993 and Nikodem, 2012).

The mean values of elastic moduli obtained from the compression test and USG measurement differ by an order of magnitude. However, the values of coefficients of correlation for BMD and Ash.D with elastic moduli fall within the range of 0.57 - 0.68. This points to the fact that, despite such large differences, the results obtained from compression test and USG are well correlated qualitatively, in spite of such large quantitative differences. The above allows to conclude that the methodics of both measurements was accepted and performed correctly. Correlation between percentage content of minerals in dry mass of the sample, and the moduli measured with an ultrasonographic method is not existent (R = 0.30 - 0.32). This likely results from the fact that this indicator refers to dry mass, and the moduli measurements were performed on wet samples. Loss of humidity after drying could have impact on the weakening of relations between the values evaluated. This impact is, however, higher for elastic moduli measured with USG method than with the compression test. The author did not find in scientific literature the descriptions of using this parameter for evaluating elastic properties of trabecular bone - perhaps due to its poor usability for that purpose.

Ash. D values obtained are at a level obtained in examinations of other authors (Mosekilde et al., 1987 and Keyak et al., 1996). Value of BMD correlation with elastic modules values were similar as the value of Ash. D. correlations with these modules. This proves the qualitative similarity of BMD and Ash.D. results obtained. BMD measurement was performed on a clinical device, according to the procedure foreseen for bone samples. With those samples, the outer layer of soft tissues surrounding the bone does not occur, as is the case with "in vivo" examinations. This procedure takes these differences into account. The samples were immersed in water of adequate depth simulating the presence of soft tissues surrounding the bone. The test parameters were also set for this type of evaluation, according to the device manufacturer's guidelines. This allows to assume that the results of actual BMD evaluations on "in vivo" patients before alloplastic implantation could give similar results. However, the author could not perform a similar verification, since the consent of the local ethics committee for the performance of tests did not permit the author of the work to access medical documentation of the persons subjected to the alloplastics treatment. It will be subject for further examination.

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