World Journal of Medical Sciences 8 (1): 48-51, 2013
ISSN 1817-3055

DOI: 10.5829/idosi.wjms.2013.8.1.65159

Change in Bone Density as a Function of Water Content

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Abstract: Water plays an important role in influencing the physical properties of biological tissues. On the basis of water content, biological tissues are differentiated as soft and hard. As compared to the rib, water content is higher in scapula and lower in femur. The paper presents a comparative account of the density of wet; oven dried and decalcified bovine bones. In the present investigation, the influence of water content on density of wet, oven dried and decalcified bovine scapula, rib and femur has been studied.

Key words: Density · Bovine Bone · Mineral · Water Content

INTRODUCTION

The composition and structure of mineral component of bone attracted the attention of physical scientists more than a century ago. Bone tissue is in a stage of continuous remodeling. While new bone is being deposited by cells called osteoblasts. The anabolic and catabolic processes account for the growth of the skeleton, for maintenance of its architecture and for the participation of bone mineral in the regulation of mineral metabolism in the body. The fibrous organic material in the bone is collagen, which belongs to the group of fibrous proteins containing while connective tissue, tendons, cartilage, etc.

The fibers of collagen are visible in the optical microscope with diameters ranging from 20 to 200μ. These fibers can be seen to be divided as primitive fibers, which have diameters in the range 2 to 10μ. Primitive fibers have thinner components, fibrils, which may be too thin (a few hundred Angstroms) to be observed under the optical microscope and hence it is necessary to use an electron microscope, Fibrils, however, are composed of still thin 'filaments' and it is the arrangement of the molecular polypeptide chains in these filaments which is revealed by X-ray diffraction studies on collagen. The X-ray diffraction pattern of crystal is unique and it is only necessary to measure the times of the pattern on an X-ray power diagram and compares the results with these from a sample of the pure substance. The difficulty with bone arises from the fact that the lines are so diffuse and that there are very few. If one bone is subjected to any treatment such as heating in order to improve the diffraction diagram, than it is not possible to be absolutely certain that no change has taken place.

Lee and Young [1] determined the composition of hard tissue proteins extracted from bovine horn, water buffalo horn rhinoceros horn to elucidate and compare the composition of the various keratin derivatives. Analysis for amino acids, monosaccharide, hexamine, uronic acids and sialic acids were found in 3 keratin derivatives and there was no difference in their composition. Marshall and Gillespie [2] examined the keratin proteins of wool, horn and hoof from sheep. The constituents of low and high sulphur proteins, isolated from 3 hard keratins of wool. Horn and hoof of sheep were compared. Horn and hoof were more similar in nature to each other than they were to wool.

The complex role of various components of bone mineralization is sufficiently exposed by various workers. A role of magnesium in the ordinary events accompanying bone mineralization has been suggested. In the adult rat, intake for 48 days of abnormal amounts of magnesium, either too much or too little, arrested the growth of the mineralization system of tibia [3].

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Smith *et al*. [4] used the computerized tomography and photon absorption has also used to determine bone cross-section or bone mass. However, these techniques also have limited sensitivity and they have not been used in monitoring fracture healing. This suggests that there is a definite need for additional means of detecting the *in vivo* condition of bone

Claudie Berger *et al*. [5] study provides a comprehensive estimate of how bone mineral density changes over time and it has important physiologic and clinical implications. They computed slops of bone mineral density measurements over time and the slope between the two points to estimate the rate of change in bone mineral density.

The density of the bone is related to mineral content, water and also the organic composition. It is obvious from the study that the deposition of calcium phosphate in the matrix of collagen is more or less homogeneous for femur, whereas in the case of scapula and rib, the deposition is inhomogeneous. However, the variation in density is less in the case of decalcified and oven dried bones [6].

MATERIALS AND METHODS

Fresh scapula, rib and femur bones of bovine were collected from the slaughter house for the present investigations. Fleshly material present on the bone is removed. Specimens were cut from the mid region of the bone. Some bones specimens were decalcified by keeping them in 9% nitric acid for 24 hours and then washed in running water for 24 hours. Fresh specimens were dried by keeping them in oven at 100°c for 24 hours.

The density of wet, decalcified and oven dried specimens was measured by taking their masses in air (m_1) and when immersed in water (m_2). Then density (d) is calculated as

$$d = \frac{m_1}{m_1 - m_2} \text{gm/cm}^3$$

Percent water content of bones ($w\%$) was determinedby taking masses before and after oven drying and using the formula

$$w\% = \frac{m_1 - m_3}{m_1} x100$$

where m_1 = mass of the bone, m_3= mass of oven dried bone.

Fractional change (df) in the density of decalcified and oven dried bones was determined by using the formula

$$f_d = \frac{d_1 - d_2}{d_1}$$

where d_1= density of wet bone, d_2= density of oven dried or decalcified bone.

For the estimation of trace elements by atomic emission spectroscopy, smooth powder of the bone samples was prepared by grinding. The powder was collected in polythene bags.

RESULTS AND DISCUSSION

Table 1, presents the data on density of wet, oven dried and decalcified bovine scapula, rib and femur bones. Standard deviation values density show the considerable variation in the case of scapula and rib, while the values are in a close range for femur.

The density of the bone is related to mineral content, water and also the organic composition. From the data on density, it is obvious that the deposition of calcium phosphate in the matrix of collagen is more or less homogenous for femur, whereas in the case of scapula and rib, the deposition is inhomogeneous. However, the variation in density is less in the case of decalcified and oven dried bones.

Fractional change in density is also presented. It can be noticed that the density of femur is more than that of rib, while in the case of scapula; it is less whatever may be the physiological condition of the bone, whether it is wet, decalcified or oven dried. The fractional change in density is high in scapula, low in rib and in between for femur, when decalcified bones are considered. In the case oven dried bones, the fractional change in density is more or less the same but it is more than that of femur.

Table 2, gives the data on water content of bovine bones – scapula, rib and femur. It is found that the water is comparatively high in scapula (12%) and low in femur (10%) when compared to the rib (11%).

Water plays an important role in influencing the physical properties of biological tissues. On the basis of water content, biological tissues are differentiated as soft and hard.

Table 3, presents a comparative account on density of wet, oven dried and decalcified bovine bones. Fractional change in density is also presented. It can be noticed that the density of femur is more than that of rib, while in the case of scapula, it is less whatever may be

Table 1: Data on density of wet, oven dried and decalcified bovine bones

		Mass of the bone (gm)			
Identifi-cation	Bone	in air		in water^3	Density (gm/cm^3)
		Wet bones			
S4A	Scapula	4.0959	1.6461	1.672	
S6T		4.4600	1.6200	1.570	
S1M		3.5512	1.2735	1.559	
S3M		3.6025	1.3805	1.621	
S4M		5.4260	2.1425	1.653	1.615±0.044
R1A	Rib	5.7500	2.2082	1.623	
R2A		4.9266	1.8963	1.625	
R1T		2.5751	1.0241	1.660	
R2T		2.8424	1.0941	1.626	
R6M		6.7900	2.4791	1.575	1.622±0.027
F3	Femur	2.2337	1.1752	2.110	
F4		3.5523	1.8442	2.080	
F2A		4.1600	2.1844	2.106	
F5A		4.4036	2.2763	2.070	
F6T		3.7521	1.9813	2.119	2.097±0.019
		Oven dried bones			
S5T	Scapula	3.7784	1.0700	1.395	
S6T		3.9672	1.1310	1.399	
S5C		1.6400	0.4650	1.396	
S6C		1.3926	0.5030	1.566	1.439±0.073
R5T	Rib	2.3234	0.2240	1.107	
R5C		1.8172	0.7170	1.652	
R6C		1.6077	0.5950	1.588	1.449±0.243
F5T	Femur	4.3608	1.9970	1.845	
F6T		3.3986	1.6730	1.970	
F5C		2.2175	1.0100	1.837	
F6C		1.2470	0.6020	1.933	1.896±0.057
		Decalcified bones			
S3C	Scapula	1.1366	0.3420	1.430	
S4C		1.6834	0.3400	1.253	1.342±0.089
R3C	Rib	1.1682	0.2360	1.253	
R4C		1.0670	0.3890	1.574	1.413 0.161
F3T	Femur	2.4512	1.1240	1.847	
F4T		2.0620	0.8800	1.744	
F3C		1.9531	0.9250	1.900	
F4C		1.6283	0.6380	1.644	1.784±0.098

Table 2: Data on water content of bovine bones

		Mass (gm)				
Identifi-cation	Bone	Wet		Oven dried	Water content (gm)	Percent water content
S1M	Scapula	3.5512	3.1118	0.4394	12.37	
S2M		4.2812	3.8065	0.4747	11.09	
S3M		3.6025	3.0895	0.5130	14.24	
S4M		5.4260	4.8796	0.5464	10.07	
S5M		3.8850	3.5352	0.3498	09.00	
S6M		4.2572	3.5292	0.7280	17.10	
S7M		2.7090	2.3419	0.3671	13.55	
S5C		1.8499	1.6400	0.2099	11.35	
S6C		1.5698	1.3926	01772	11.29	
S5T		4.2902	3.7784	0.5118	11.92	
S6T		4.4600	3.9672	0.4928	11.05	12.09±2.11

Table 2: Continue

		Mass (gm)				
Identifi-cation	Bone	Wet		Oven dried	Water content (gm)	Percent water content
R1M	Rib	6.7900	6.0393	0.7507	11.06	
R2M		10.9562	9.4230	1.5332	13.99	
R3M		8.8540	7.8511	1.0029	11.33	
R5C		2.0535	1.8172	0.2363	11.51	
R6C		1.8100	1.6077	0.2023	11.18	
R5T		2.9523	2.6064	0.3459	11.71	
R6T		1.9042	1.6920	0.2122	11.14	11.70±0.91
F5C	Femur	2.4862	2.2175	0.2687	10.81	
F6C		1.3798	1.2470	0.1328	09.62	
F5T		4.8393	4.3608	0.4786	09.89	
F6T		3.7521	3.3986	0.3535	09.42	9.94±0.53

Table 3: A comparison of density of wet, oven dried and decalcified bovine scapula, rib and femur bones

	Density (gm/cm^3)			Fractional change in density	
Bone	Wet	Decalcified	Oven dried	Decalcified	Oven dried
Scapula	1.615±0.044	1.342±0.089	1.439±0.073	0.169	0.109
Rib	1.622±0.027	1.413±0.161	1.449±0.243	0.129	0.107
Femur	2.097±0.019	1.784±0.098	1.896±0.057	0.149	0.096

the physiological condition of the bone, whether it is wet, decalcified or oven dried, The fractional change in density is high in scapula, low in rib and in between for femur, when decalcified bones are considered. In the case oven dried bones, the fractional change in density is more or less the same but it is more than that of femur.

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