

The living pattern of Awo Omamans will rapidly change as community development progresses. The effect of these changes on the decay rate should be closely watched, preferably after a more detailed dental survey of the villagers has been made.

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References and Notes

1. This work was performed while I was serving as medical field consultant for the Unitarian Service Committee, Village Improvement Project, Awo Omamma, Nigeria.
2. W. J. Pelton and E. H. Pennell, *J. Am. Dental Assoc.* 49, 439 (1954).
3. Determinations were obtained with the assistance of Shinji Soneda, sanitary engineer, Hawaii State Dept. of Health, Hilo.
4. Determinations were obtained with the assistance of Dr. Bruce Forsyth, Regional Dental Consultant, U.S. Public Health Service, and Dr. Manuel C. W. Kau, executive officer, Dental Health Div., Hawaii State Dept. of Health.
5. Water and tooth specimens were obtained by Dr. Hazel M. Hauck, professor of nutrition, Cornell University, Ithaca, N.Y., during her residence in Awo Omamma.
6. O. Nzekwu, *Nigeria Magazine* 71, 298 (1961).
7. Many other varieties of chewing stick are in use. Commonly found are: Calabar, and in the Ibo language, Atu-ofe, Atu-uloro, Mbacara, Uda, Atu-nkperere, Atu-ubere, Mbeji, Nkpaku, Uguri, and Nkpa.

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Bone Density Measurements in vivo

Abstract. Measurements in vivo are complicated by physical and physiological problems. Lack of standard techniques between laboratories and confusion in terminology have prompted this report. The theory underlying the method is presented with data comparing values obtained by different methods.

One method of measuring bone density is comparison of the absorption of x-radiation by a test object with a reference object. When x-rays are absorbed by a uniform material the intensity of radiation is reduced by a constant fraction per unit length of path through the material (*linear absorption coefficient* or μ). An equivalent definition for μ is obtained when a beam of x-rays of unit area in cross section traverses unit volume of the substance. The fraction of energy absorbed when a beam of unit cross section traverses a unit mass of material is called the *mass absorption coefficient* (μ/ρ , where ρ is the density of the material). It is the same for substances of the same atomic number.

When an x-ray beam penetrates to a depth t of a reference object, the intensity is $I = I_0 e^{-\mu t}$ where I_0 is the intensity at the surface. For a test ob-

ject, this may be expressed as $I' = I_0 e^{-\mu' t'}$.

If masses of the reference object and the test object absorb equal fractions of the total energy, then

$$\mu t = \mu' t'$$

or

$$\frac{\mu'}{\mu} = \frac{t}{t'}$$

If the reference object has the same mass absorption coefficient (μ/ρ) as the test object (μ'/ρ'), then

$$\frac{\mu'}{\mu} = \frac{\rho'}{\rho} \quad (1)$$

therefore

$$\frac{\mu'}{\mu} = \frac{\rho'}{\rho} = \frac{t}{t'}$$

and

$$\rho' = \frac{\rho t}{t'} \quad (2)$$

In this laboratory, an x-ray picture is taken simultaneously of the little finger and of a reference wedge of known density (ρ). The wedge is a homogeneous alloy of 92.8 percent aluminum and 7.2 percent zinc. Its effective atomic number approximates that of hydroxyapatite (16.65) (I). Its mass absorption coefficient, thus, is closely equivalent to that of bone mineral (see Eq. 1), making it appropriate for comparing bones of different density.

After trying various exposure times, the most suitable was chosen along the linear portion of the film response curve. The standard conditions used are 50 kv, 10 ma, 1 second, and 36 inches focal distance.

A recording microphotometer scans across the film image of phalanx 5-2 at its center and along the central longitudinal axis of the wedge image (Fig. 1). Since x-rays and light are absorbed exponentially, the slope of the wedge trace differs from the slope of the bone trace (0.1). Measured with a planimeter, the areas under the finger trace represent integrated mass absorption due to the mass of bone plus over- and underlying flesh ($C + B + C'$) and due to flesh lateral to the bone ($A + A'$). Dividing by corresponding pathways of the x-ray beam, a height is found representing the average absorption for each portion. These heights locate places on the base line of the wedge trace indicating thicknesses of wedge which absorb in the same amount.

Average thicknesses of flesh $A + A'$ and of $C + C'$ are calculated from the cross-sectional areas determined from measurements on the trace (r , x , and length of pathways) assuming circular outlines for flesh and bone. Equations used to determine areas are

$$A = \frac{\pi r^2}{2} - [x(r^2 - x^2)^{1/2} + r^2 \sin^{-1}(x/r)]$$

(with the angle expressed in radians)

and

$$C + C' = \pi r^2 - [\pi x^2 + \text{area } 2A]$$

A ratio of the thicknesses is used to correct the wedge thickness representing flesh $A + A'$ (Fig. 1, wedge trace) to that representing flesh $C + C'$. The equivalent wedge thickness for flesh plus bone minus that for flesh $C + C'$ gives the thickness (t) of wedge of known density which absorbs the same amount of energy as the hydroxyapatite in the bone slice of unknown density.

A bone consists of supporting tissues plus hydroxyapatite. The density of bone is required—not that of hydroxyapatite. The average thickness (t') of the bone penetrated by the x-ray is obtained from linear measurements on the trace.

The measured values for ρ , t , and t' are substituted in Eq. 2 to solve for ρ' .

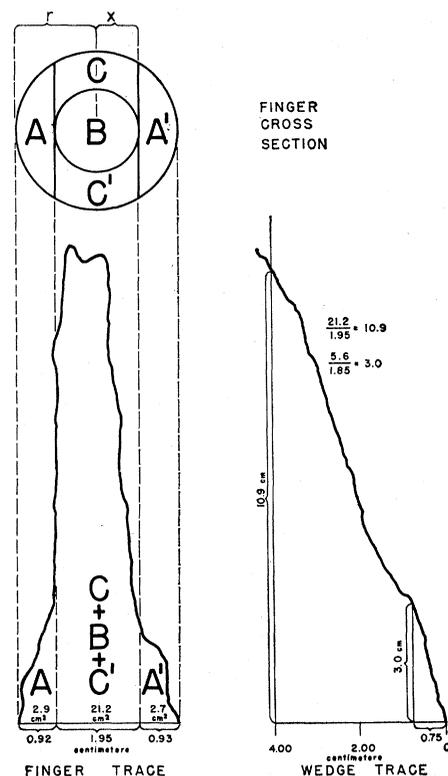


Fig. 1. Measurements needed to calculate density values.

Table 1. Comparison of measurements for three girls and six adults (five women and one man).

Measurement	Children	Adults
<i>Bone density measurements of phalanx 5-2</i>		
Circle	0.72 ± 0.03	0.89 ± 0.08
Ellipse	0.77 ± 0.08	1.18 ± 0.11
<i>Axes measurements (from trace)</i>		
Diameter or a-axis (cm)		
finger	3.18 ± 0.09	3.81 ± 0.53
bone	1.38 ± 0.07	1.76 ± 0.27
b-axis (cm)		
finger	3.31 ± 0.07	3.59 ± 0.63
bone	1.02 ± 0.08	1.14 ± 0.17

The bone density value is expressed as x-ray equivalent grams of alloy per cubic centimeter of bone.

Certain approximations for in vivo determinations must be made, for example, the elimination of the effect of soft tissue. The wedge is made with a mass absorption characteristic similar to bone mineral but it cannot, also, absorb like flesh. Omnell (1) shows that mass absorption coefficients change with change in energy level and with substances of different atomic number. Rowland *et al.* (2), however, showed that most of the absorption by bone of x-rays with low energies (4 to 10 kv) is due to mineral components and that contributions due to soft tissues may be neglected. In the Tennessee studies the energy level (50 kv) is fairly low and the phalanx bone has only a small amount of tissue. Although a correction for surrounding tissue is made, one cannot be made for the soft tissue within the bone. The difference from bone to bone per cubic centimeter can be considered negligible or may warrant the use of the term, bone density index but not bone density coefficient.

The degree of approximation to the actual density value depends, in part, upon the extent of deviation from the assumed circular outline. With lateral and anterior-posterior views of the finger, the cross-sectional areas can be determined as ellipses with two measured axes rather than as circles with one.

In children, 7 to 8 years old, density values (Table 1) for phalanx 5-2 were essentially the same, whether calculated from circular or elliptical outlines, making one radiation exposure seem adequate.

For adults, the deviation of the phalanx outline from a circle is greater than in a child as shown by the b-axis measurements in Table 1. A greater deviation of the density value results. The precision desired must be weighed

against the disadvantages of taking two x-ray pictures.

Bone densities of phalanx 5-2 in vitro (five men, 1.03 ± 0.09; five women, 0.93 ± 0.24) were obtained by Virtama *et al.* (3) from weight per volume measurements. Rowland *et al.* (2) expressed bone density for 22 subjects in terms of hydroxyapatite (1.19 ± 0.12) for the human femur in vitro. Considering unknown ages and nutritional histories, these values are in good agreement with the in vivo phalanx 5-2 values for adults (Table 1) and with previous findings of Williams and Samson (4) for Orientals (eight men, 1.00 ± 0.20; eight women, 1.08 ± 0.16) and Americans (eight men, 1.13 ± 0.27; eight women, 1.12 ± 0.21).

For useful bone density measurements, standard techniques and exact terminology must be observed. Work is continuing here to develop an instrument for greater precision and rapidity of measurement (5).

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References and Notes

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4. D. E. Williams and A. Samson, *J. Am. Dietet. Assoc.* **36**, 462 (1960).
5. Grateful acknowledgment is made of the help from Dr. Isabel Tipton, Physics Department, University of Tennessee. This research was supported, in part, by grant A-3345 from the U.S. Public Health Service.

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Heart Autotransplantation: Effect on Myocardial Catecholamine and Histamine

Abstract. Complete excision and reimplantation of the canine heart is followed by a fall in myocardial norepinephrine to negligible levels. These decreases are attributable to the sympathetic denervation which necessarily accompanies the operative procedure. Myocardial histamine levels in survivors of this operation were not significantly different from those of normal dogs.

Complete excision and reimplantation of the dog's heart in its normal site is associated, in our hands, with an immediate mortality of 68 percent (1). In surviving animals we have observed certain alterations in cardiac function, presumably related to the

Table 1. Myocardial catecholamines and histamine in normal dogs and dogs after orthotopic cardiac autotransplantation.

Sample No.	Assay (μg/g)	
	Catecholamines	Histamines
<i>Normals</i>		
C 4	0.88	2.15
C 7	0.80	3.28
C 9	0.72	2.27
C 11	0.88	4.10
C 12	0.73	7.04
C 14	0.45	5.50
C 16	1.00	5.80
C 18	0.84	6.50
Average	0.79 ± 0.16*	4.58 ± 1.90*
<i>Autotransplants</i>		
TR 3	0.01	4.50
TR 5	0.05	3.60
TR 6	0.00	6.62
TR 11	0.00	4.62
TR 19	0.01	4.65
Average	0.01 ± 0.02*	4.80 ± 1.12*

* Standard deviation

complete extrinsic denervation which necessarily accompanies this procedure (2). Extrinsic sympathetic denervation should be associated with depletion of catecholamine stores in these hearts (3). Moreover, the possibility of histamine depletion has been raised by reports that denervation of other organs resulted in loss of tissue histamine. We now present the results of histamine and catecholamine assay in orthotopic autotransplanted canine myocardial tissues.

The cardiac autotransplantation involves division and reanastomosis of the two venae cavae, the ascending aorta, and the main pulmonary artery. Temporary separation of the heart from the mediastinum with complete extrinsic denervation is completed by cutting the left atrium away from its posterior attachment and resuturing it in its original location.

Five dogs which survived the cardiac autotransplantation procedure were studied from 7 to 28 days after operation. Immediately after sacrifice of the animal, the heart was excised and frozen at the temperature of dry ice. The entire heart, including atria and ventricles, was minced and thoroughly mixed. Samples of the mixture were subsequently studied. Catecholamines were assayed fluorometrically by the method of Crout *et al.* (4). Oxidations were carried out at pH 4.9 in order to express results as norepinephrine equivalents. Tissue histamine was assayed fluorometrically by the method of Shore and his co-workers (5). Eight normal dogs of comparable size were sacrificed in a similar manner. Assay