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D. D. Thompson,[1] *Ph.D.*

The Core Technique in the Determination of Age at Death in Skeletons

Determining age at death beyond 50 years in skeletons has posed problems for physical anthropologists, forensic scientists, and archeologists. Morphological aging methods such as pubic symphyseal remodeling [*1-5*], cranial suture closure [*1,2,6,7*], and the degree of osteoarthritis [*8*] are often inaccurate or not appropriate in aging skeletons of persons older than 50 years. Histological methods of estimating age at death in skeletons [*9-13*], overcoming many of the subjective criteria associated with morphological aging methods, are receiving increasing attention for their ability to age skeletons accurately from birth to old age. Of the histological methods that use cortical bone samples, Kerley's method [*10*] has been shown to be the most accurate. Current histological methods, however, have shortcomings that limit their widespread application by physical anthropologists and forensic scientists. The principal shortcoming is the need for complete cross sections of diaphyseal bone. With Ubelaker's recent finding [*14*] that age-related histological changes in bone may be population-specific, the need for a nondestructive technique of bone sample acquisition becomes important. To confirm or reject the findings that populations may vary in their rates of osteon turnover, thereby affecting age estimations obtained by histological methods, it is necessary to acquire bone samples from large skeletal series of known age at death. Access to these skeletons as well as forensically derived skeletons depends on a technique that minimizes the physical damage to a skeleton.

The purposes of this study were (1) to propose a histological method that uses a small core of cortical bone to estimate age at death, primarily beyond 50 years of age, in skeletons; (2) to provide an objective method for quantifying cortical bone microstructures used in age estimation; and (3) to examine the feasibility of obtaining estimates of age at death from bones of the upper and lower extremities instead of the lower extremities only.

Materials and Methods

Sample Description

The sample used in this study consisted of 116 human cadavers—64 males and 52 females. Age at death in years for each cadaver was obtained from death certificates. Males ranged in age from 30 to 97 years with a mean of 71.48 years (standard deviation $SD = 12.90$) and females from 43 to 94 years with a mean of 71.94 years ($SD = 13.81$). The primary cause of death was known for each individual. In some cases more complete

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[1]Assistant professor in residence, Laboratory of Biological Anthropology, Department of Biobehavioral Sciences, University of Connecticut, Storrs.

medical histories were available that indicated secondary causes of death and other chronic conditions affecting the individual prior to death.

Each cadaver was categorized into either a nonpathological or a pathological group with respect to the primary cause of death. A nonpathological categorization denoted a cause of death that had no apparent affect on cortical bone remodeling in the person's lifetime whereas a pathological categorization indicated a cause of death that has been shown to affect cortical bone remodeling prior to death, for example, from renal insufficiency [*15-17*] or diabetes mellitus [*18,19*]. Other factors that may have further influenced cortical bone remodeling, such as the length of time persons were confined to bedrest, medications administered, parity, and duration of illness prior to death, were not available to the investigator. Sample sizes and age distributions for the entire series and then the nonpathological group presented according to sex and bones sampled are summarized in Table 1. The pathological group was not analyzed separately because of the heterogeneous composition of the group with respect to the primary causes of death.

Core Technique

Specially constructed bone corers were used to obtain diaphyseal cortical bone samples from the cadavers. The corers, mounted in an high-speed Dremel drill, removed bone cores 0.4 cm in diameter. The principal bones from which cores were removed included the right and left femur and tibia. In certain cases one bone was missing from a cadaver as a result of postmortem amputation. This resulted in uneven sample sizes for the bones that were cored. A total of 429 bone cores was removed from the femurs and tibiae of the 116 cadavers. Additionally, a total of 122 bone cores was removed from right and left humeri and ulnas in 31 individuals. One bone core was removed from each of the bones. The total sample sizes for each bone are presented in Table 1. The bone cores were removed from the following locations on each of the bones: femur, midshaft, anterior surface; tibia, midshaft, medial surface; humerus, midshaft, medial to deltoid tuberosity; and ulna, one third of the distance from the distal end, lateral surface.

Description of Variables

Nineteen variables were ascertained from each bone core. Each variable used in this study has been shown to vary as a function of age [*20*]. While other studies have relied exclusively on histological variables to estimate age at death, this study was designed to examine the feasibility of including variables other than those histologically derived, such as cortical thickness, bone density, and bone mineral content.

Cortical thickness of the bone (measured from the core) was determined to the nearest 0.05 mm after adherent marrow and periosteum had been removed. The cores were ground at their endosteal end with #800 carborundum paper to yield a cylinder as nearly perfect as possible. Wet bone density (g/cm^3) was estimated from the bone core by dividing the volume of the core by the wet weight of the core. The core weight was determined to the nearest 0.0001 g by using a Mettler Model H207 balance. Core diameter and core length were determined to the nearest 0.05 mm.

The mineral content of each core was measured by ^{125}I photon absorptiometric analysis (Norland-Cameron Bone Mineral Analyzer). The bone mineral index (g/cm^2) was found by dividing the mineral content (g/cm) by the core length (cm). The mineral content (g/cm) was read directly from the Bone Mineral Analyzer and the core length was measured with calipers as well as read directly from the Bone Mineral Analyzer. The principles of photon absorptiometric analysis used in the determination of bone mineral have been summarized by Cameron and Sorenson [*21*]. During scanning each core was submerged

TABLE 1—*Sample sizes and age distributions for the groups used in this study.*

Group	Bone[a]	*n*	Mean Age, years	*SD*	Coefficient of Variation
Whole series	LF	91	69.47	13.56	0.20
	RF	113	72.10	12.76	0.18
	LT	112	71.64	13.33	0.19
	RT	113	72.06	12.94	0.19
All males	LF	53	69.94	13.20	0.19
	RF	63	72.67	11.85	0.16
	LT	62	71.87	12.97	0.18
	RT	63	72.24	11.95	0.17
All females	LF	38	68.82	14.21	0.21
	RF	50	71.38	13.91	0.19
	LT	50	71.36	13.89	0.19
	RT	50	71.84	14.21	0.20
Nonpathological samples	LF	68	70.68	13.23	0.19
	RF	90	73.20	12.80	0.17
	LT	89	72.90	12.95	0.18
	RT	90	73.16	13.03	0.18
Nonpathological males	LF	41	71.49	12.13	0.17
	RF	54	73.93	11.72	0.16
	LT	53	73.45	12.01	0.16
	RT	53	73.43	12.01	0.16
Nonpathological females	LF	27	69.44	14.91	0.21
	RF	36	72.11	14.38	0.20
	LT	36	72.08	14.36	0.20
	RT	37	72.76	14.54	0.20
Upper extremities	LH	29	67.69	13.55	0.20
	RH	31	68.00	13.60	0.20
	LU	31	68.26	13.98	0.20
	RU	31	69.42	13.74	0.20

[a] LF = left femur; RF = right femur; LT = left tibia; RT = right tibia; LH = left humerus; RH = right humerus; LU = left ulna, and RU = right ulna.

in 3 cm of water in a Plexiglas® box and scanned from the periosteal margin to the endosteal margin. Scanner speed was 1 mm/s and the scan beam was collimated to 1.5 mm. Four scans were made on each core and the mean of the scans was computed. After being scanned each core was sectioned with a Buehler Isomet® saw. A section approximately 90 μm in thickness was removed from each core in a plane that was transverse to the longitudinal axis of the long bone, ground to a thickness of 80 μm, and ultrasonically cleaned. The prepared bone sections were then mounted on microscope slides with synthetic resin mountant.

Microscopic Examination of Cortical Bone Microstructures

Microscopic analysis of the bone sections was done with a phase contrast microscope at $\times 100$. Measurements on secondary osteons and Haversian canals (including primary osteons) in the bone sections were achieved by using stereological procedures of morphometry outlined by Elias and Pauly [*22*], Frost [*23*], and Weibel [*24*]. A 10 by 10 grid eyepiece disk micrometer (measuring 0.992 mm^2 at $\times 100$) was used in the stereological quantification of secondary osteons and Haversian canals. Three stereological principles were employed.

First, the aggregate areas of secondary osteon lamellae and Haversian canals were each assessed and then summed together to yield the areal surface of a field containing osteons. Aggregate osteon lamellae and Haversian canal areas were determined in four adjacent periosteal fields by using the point count method described by Frost [*23*].

Second, the number of secondary osteons and Haversian canals was determined in the same four adjacent periosteal fields. Primary osteons were included in Haversian canal counts but not in secondary osteon counts. Osteons and Haversian canals bisected by the grid's outer perimeter and thus only partially contained in the grid were counted on an alternate basis. The first such structure would be counted while the second would be excluded. This procedure was continued until all structures were accounted for in the fields.

Third, the aggregate perimeters B_A of secondary osteons and Haversian canals were quantified with the formula [*24*]

$$B_A = (\pi/2)\, I_L$$

where I_L is the number of intersections of a structure's perimeter per unit length of test line. In this study each grid line measured 0.992 mm and the 10 by 10 grid contained 22 test lines. The total length of test line was 21.82 mm. The value of I_L was thus computed by totaling the number of line intersections of first the secondary osteon reversal lines and then the Haversian canal perimeters and dividing each number by 21.82 mm. Each I_L was next multiplied by $\pi/2$ to yield the aggregate perimeters for secondary osteons and Haversian canals (including primary osteons) in each of the four periosteal fields. Mean aggregate perimeters, areas, and numbers were computed for secondary osteons and Haversian canals for the four fields and used in all further analyses. Additionally, individual secondary osteon and Haversian canal areas and perimeters were estimated by dividing the mean aggregate areas and perimeters for each section by the respective mean number of secondary osteons or Haversian canals.

Three ratios were also derived from these microstructural quantifications. Ratio 1 was found by dividing the mean aggregate Haversian area by the mean aggregate secondary osteon lamellae area for each section; Ratio 2 was found by dividing the mean aggregate Haversian canal perimeter by the mean aggregate secondary osteon perimeter; and Ratio 3 was found by dividing the individual Haversian canal perimeter by the individual secondary osteon perimeter for each section. The 19 variables are summarized in Table 2.

With age as the dependent variable, the 19 variables derived from each core were subjected to stepwise linear regression analysis [*25*] to select the variable or combination of variables that would estimate age at death in skeletons with the lowest standard error of the estimate *SEE* and the highest coefficient of determination. Twenty-eight separate regression analyses were performed with the data collected from the 116 cadavers (Table 3). Inclusion of variables in an equation was based on the multiple correlation coefficient.

The regression equations are presented in their entirety so that future investigators can select the equations best fitting their sample to be aged. The regression equation to be used depends on the information available from the skeleton and the variables collected. An archeologically derived skeleton would generally be accurately designated either male or female, but the cause of death would be lacking. The appropriate equation in the estimation of age in this skeleton would be the one lumping all males (Table 3, Analyses 5 to 8, depending on the bone used) or all females (Table 3, Analyses 9 to 12, depending on the bone used).

Results

Stepwise linear regression analysis revealed one variable, the osteon area, to estimate age at death consistently in this series with the greatest accuracy (Table 3). Of the 28

TABLE 2—*Summary of the 19 variables and their abbreviations used in the regression analyses.*

Variable	Abbreviation	Variable Description
1. cortical thickness, mm	CTHICK	measured from intact core with calipers
2. core weight, g	COREWT	wet weight of refinished core
3. cortical bone density, g/cm^3	CDEN	weight of core per unit volume of core
4. mineral content, g/cm	CMC	measured in cores by Bone Mineral Analyzer
5. mineral index, g/cm^2	CMCC	mineral content/refinished core length
6. aggregate osteon lamellae area, %	OSTA	percentage of area of fields containing osteon lamellae
7. aggregate Haversian canal area, %	HCA	percentage of area of fields containing canals
8. osteon area, %	OSTHC	aggregate osteon lamellae plus Haversian canal area
9. secondary osteon number	NUMOST	number of secondary osteons in a field
10. Haversian canal number	NUMHC	number of canals and primary osteons in a field
11. individual osteon lamellae area, %	INDOSTA	osteon lamellae area/secondary osteon number
12. individual Haversian canal area, %	INDHCA	Haversian canal area/Haversian canal number
13. aggregate osteon perimeter, mm	OSTBA	total osteon perimeter length in a field
14. aggregate Haversian canal perimeter, mm	HCBA	total Haversian canal perimeter length in a field
15. individual osteon perimeter, mm	IOSTBA	aggregate osteon perimeter/osteon number
16. individual Haversian canal perimeter, mm	IHCBA	aggregate canal perimeter/canal number
17. Ratio 1	RATIOA	aggregate Haversian canal area/aggregate secondary osteon lamellae area
18. Ratio 2	RATIOB	aggregate Haversian canal perimeter/aggregate osteon perimeter
19. Ratio 3	RATIOC	individual canal perimeter/individual osteon perimeter

TABLE 3—*Stepwise regression analysis of the 28 groups used in this study.*

Analysis Number	Group	Bone	n	Step Number	Variable Entered	Regression Equation	Multiple Correlation Coefficient	Coefficient of Determination	Standard Error of Estimate, years
1	whole series	left femur	91	1	OSTHC	$y = 6.677 + 101.936x_1$	0.7734	0.5982	8.6455
				2	CTHICK	$y = 20.969 + 95.278x_1 - 2.314x_2$	0.8063	0.6502	8.1124
				3	IOSTBA	$y = 47.644 + 96.394x_1 - 2.457x_2 - 47.590x_3$	0.8276	0.6849	7.7438
				4	OSTBA	$y = 72.059 + 127.853x_1 - 1.797x_2 - 83.949x_3 - 2.739x_4$	0.8551	0.7312	7.1929
				5	NUMOST	$y = 28.978 + 128.557x_1 - 1.796x_2 - 7.543x_3 - 7.633x_4 + 2.688x_5$	0.8624	0.7437	7.0651
2	whole series	right femur	113	1	OSTHC	$y = 12.409 + 91.936x_1$	0.7887	0.6221	7.8789
				2	IOSTBA	$y = 30.473 + 94.172x_1 - 34.688x_2$	0.8014	0.6422	7.7007
				3	CTHICK	$y = 42.175 + 91.588x_1 - 41.134x_2 - 1.399x_3$	0.8147	0.6638	7.4992
				4	OSTBA	$y = 52.063 + 102.082x_1 - 54.796x_2 - 1.183x_3 - 1.003x_4$	0.8196	0.6718	7.4438
				5	NUMOST	$y = 35.747 + 100.985x_1 - 26.752x_2 - 1.194x_3 - 2.791x_4 + 1.058x_5$	0.8250	0.6807	7.3760
3	whole series	left tibia	112	1	OSTHC	$y = 20.835 + 82.235x_1$	0.7036	0.4950	9.5163
				2	IOSTBA	$y = 45.616 + 88.260x_1 - 51.541x_2$	0.7351	0.5403	9.1209
				3	NUMOST	$y = 94.199 + 130.361x_1 - 137.057x_2 - 1.549x_3$	0.7714	0.5950	8.6008
				4	CMC	$y = 100.361 + 118.566x_1 - 130.198x_2 - 1.296x_3 - 54.397x_4$	0.7857	0.6174	8.3989
4	whole series	right tibia	113	1	OSTHC	$y = 20.632 + 82.475x_1$	0.7441	0.5537	8.6822
				2	IOSTBA	$y = 42.986 + 88.917x_1 - 47.830x_2$	0.7707	0.5939	8.3187
				3	OSTBA	$y = 73.750 + 129.529x_1 - 94.988x_2 - 3.146x_3$	0.8061	0.6498	7.7603
				4	CMCC	$y = 104.964 + 120.319x_1 - 95.279x_2 - 3.000x_3 - 68.935x_4$	0.8180	0.6692	7.5778

TABLE 3—*Continued.*

Analysis Number	Group	Bone	*n*	Step Number	Variable Entered	Regression Equation	Multiple Correlation Coefficient	Coefficient of Determination	Standard Error of Estimate, years
5	all males	left femur	53	1	OSTHC	$y = 8.387 + 100.133x_1$	0.7873	0.6199	8.2167
				2	CTHICK	$y = 25.014 + 93.735x_1 - 2.610x_2$	0.8199	0.6723	7.7058
				3	IOSTBA	$y = 53.989 + 95.112x_1 - 2.922x_2 - 51.114x_3$	0.8467	0.7168	7.2354
				4	OSTBA	$y = 73.137 + 120.584x_1 - 2.619x_2 - 80.433x_3 - 2.070x_4$	0.8586	0.7373	7.0418
				5	NUMOST	$y = 20.732 + 116.813x_1 - 2.501x_2 + 12.810x_3 - 7.735x_4 + 3.031x_5$	0.8699	0.7567	6.8479
6	all males	right femur	63	1	OSTHC	$y = 18.413 + 84.646x_1$	0.8061	0.6499	7.0675
				2	IOSTBA	$y = 39.056 + 89.825x_1 - 42.502x_2$	0.8303	0.6894	6.7118
				3	CTHICK	$y = 50.783 + 90.256x_1 - 49.121x_2 - 1.680x_3$	0.8461	0.7159	6.4733
				4	CDEN	$y = 82.772 + 90.273x_1 - 53.212x_2 - 1.558x_3 - 16.403x_4$	0.8523	0.7265	6.4061
7	all males	left tibia	62	1	OSTHC	$y = 19.450 + 84.929x_1$	0.7054	0.4977	9.2687
				2	IOSTBA	$y = 43.351 + 89.082x_1 - 49.070x_2$	0.7338	0.5384	8.9594
				3	NUMOST	$y = 89.861 + 132.473x_1 - 132.573x_2 - 1.515x_3$	0.7617	0.5802	8.6178
				4	CMC	$y = 102.007 + 126.336x_1 - 136.646x_2 - 1.467x_3 - 48.648x_4$	0.7738	0.5988	8.4985
				5	COREWT	$y = 106.027 + 125.897x_1 - 141.415x_2 - 1.596x_3 - 163.603x_4 + 211.389x_5$	0.7847	0.6157	8.3916
8	all males	right tibia	63	1	OSTHC	$y = 24.982 + 77.260x_1$	0.7126	0.5078	8.4488
				2	RATIOA	$y = 17.383 + 75.131x_1 + 45.816x_2$	0.7463	0.5570	8.0821
				3	IOSTBA	$y = 38.002 + 81.605x_1 + 49.204x_2 - 46.756x_3$	0.7705	0.5936	7.8063
				4	HCBA	$y = 53.997 + 116.335x_1 + 84.131x_2 - 79.992x_3 - 6.612x_4$	0.8010	0.6416	7.3994

				5	CMCC	$y = 81.153 + 115.322x_1 + 75.997x_2 - 82.491x_3 - 7.136x_4 - 55.734x_5$	0.8087	0.6539	7.3293
9	all females	left femur	38	1	OSTHC	$y = 4.097 + 104.755x_1$	0.7597	0.5771	9.3665
				2	CTHICK	$y = 24.239 + 93.309x_1 - 3.475x_2$	0.8255	0.6814	8.2451
				3	OSTBA	$y = 27.727 + 112.093x_1 - 2.829x_2 - 1.851x_3$	0.8450	0.7140	7.9250
				4	IOSTBA	$y = 66.568 + 126.957x_1 - 1.978x_2 - 3.077x_3 - 69.156x_4$	0.8693	0.7557	7.4356
				5	INDOSTA	$y = 82.386 + 118.387x_1 - 1.485x_2 - 2.552x_3 - 139.660x_4 + 751.730x_5$	0.8795	0.7734	7.2712
10	all females	right femur	50	1	OSTHC	$y = 1.829 + 105.431x_1$	0.7976	0.6361	8.4788
				2	CMC	$y = 24.721 + 87.001x_1 - 84.051x_2$	0.8445	0.7131	7.6086
				3	COREWT	$y = 24.792 + 84.664x_1 - 246.216x_2 - 298.093x_3$	0.8585	0.7370	7.3641
				4	HCBA	$y = 27.255 + 93.251x_1 - 248.505x_2 + 300.873x_3 - 2.010x_4$	0.8641	0.7467	7.3062
				5	RATIOB	$y = 15.846 + 104.218x_1 - 246.828x_2 + 326.837x_3 - 4.047x_4 + 24.388x_5$	0.8702	0.7572	7.2236
11	all females	left tibia	50	1	OSTHC	$y = 22.075 + 79.674x_1$	0.7029	0.4940	9.9859
				2	CTHICK	$y = 36.611 + 76.231x_1 - 3.975x_2$	0.7712	0.5948	9.0309
				3	IOSTBA	$y = 54.718 + 82.759x_1 - 3.471x_2 - 41.605x_3$	0.7861	0.6179	8.8642
				4	NUMOST	$y = 88.248 + 113.313x_1 - 2.617x_2 - 105.021x_3 - 1.159x_4$	0.8052	0.6484	8.5973
12	all females	right tibia	50	1	OSTHC	$y = 14.169 + 90.306x_1$	0.7877	0.6205	8.8445
				2	IOSTBA	$y = 37.308 + 95.174x_1 - 46.375x_2$	0.8103	0.6566	8.5026
				3	NUMHC	$y = 123.205 + 141.522x_1 - 180.706x_2 - 2.238x_3$	0.8649	0.7481	7.3608
				4	CTHICK	$y = 131.231 + 123.466x_1 - 170.266x_2 - 1.837x_3 - 3.082x_4$	0.8822	0.7782	6.9831
13	nonpathological group	left femur	68	1	OSTHC	$y = 8.169 + 100.523x_1$	0.7684	0.5904	8.5335
				2	IOSTBA	$y = 40.643 + 101.645x_1 - 58.800x_2$	0.8035	0.6456	7.9984
				3	OSTBA	$y = 72.761 + 131.471x_1 - 97.270x_2 - 3.031x_3$	0.8453	0.7145	7.2349
				4	CMC	$y = 82.223 + 118.533x_1 - 97.736x_2 - 2.469x_3 - 41.586x_4$	0.8589	0.7376	6.9905

TABLE 3—*Continued.*

Analysis Number	Group	Bone	n	Step Number	Variable Entered	Regression Equation	Multiple Correlation Coefficient	Coefficient of Determination	Standard Error of Estimate, years
14	nonpathological group	right femur	90	1	OSTHC	$y = 13.271 + 91.028x_1$	0.7879	0.6208	7.9270
				2	IOSTBA	$y = 37.943 + 94.930x_1 - 47.458x_2$	0.8098	0.6558	7.5963
				3	CTHICK	$y = 48.416 + 92.826x_1 - 52.250x_2 - 1.446x_3$	0.8238	0.6787	7.3813
				4	OSTBA	$y = 57.761 + 102.746x_1 - 64.648x_2 - 1.169x_3 - 1.014x_4$	0.8285	0.6865	7.3345
15	nonpathological group	left tibia	89	1	OSTHC	$y = 23.277 + 79.089x_1$	0.7243	0.5246	8.9768
				2	IOSTBA	$y = 49.582 + 87.092x_1 - 56.225x_2$	0.7622	0.5810	8.4764
				3	CTHICK	$y = 57.423 + 85.044x_1 - 56.176x_2 - 1.816x_3$	0.7815	0.6107	8.2182
				4	NUMOST	$y = 80.431 + 107.697x_1 - 99.519x_2 - 1.449x_3 - 0.822x_4$	0.7909	0.6256	8.1076
16	nonpathological group	right tibia	90	1	OSTHC	$y = 21.129 + 82.517x_1$	0.7597	0.5771	8.5209
				2	IOSTBA	$y = 47.746 + 91.086x_1 - 57.471x_2$	0.7958	0.6333	7.9796
				3	NUMHC	$y = 98.576 + 118.566x_1 - 135.701x_2 - 1.352x_3$	0.8197	0.6720	7.5914
				4	CMCC	$y = 132.876 + 113.857x_1 - 144.623x_2 - 1.441x_3 - 64.463x_4$	0.8298	0.6886	7.4391
17	nonpathological males	left femur	41	1	OSTHC	$y = 12.207 + 95.775x_1$	0.7799	0.6083	7.6888
				2	CTHICK	$y = 24.399 + 94.460x_1 - 2.367x_2$	0.8106	0.6571	7.2883
				3	IOSTBA	$y = 55.167 + 91.446x_1 - 2.705x_2 - 49.406x_3$	0.8404	0.7063	6.8359
				4	OSTBA	$y = 75.209 + 113.167x_1 - 2.470x_2 - 78.530x_3 - 1.899x_4$	0.8532	0.7280	6.6686
18	nonpathological males	right femur	54	1	OSTHC	$y = 21.450 + 80.749x_1$	0.7966	0.6346	7.1509
				2	IOSTBA	$y = 43.071 + 85.641x_1 - 43.766x_2$	0.8221	0.6758	6.8017
				3	CTHICK	$y = 55.007 + 87.496x_1 - 50.457x_2 - 1.920x_3$	0.8429	0.7105	6.4914

				4	RATIOA	$y = 52.905 + 84.523x_1 - 51.962x_2 - 1.530x_3 + 15.421x_4$	0.8504	0.7232	6.4119
19	nonpathological males	left tibia	53	1	OSTHC	$y = 25.966 + 75.930x_1$	0.6776	0.4592	8.9151
				2	IOSTBA	$y = 49.829 + 82.248x_1 - 51.346x_2$	0.7125	0.5077	8.5909
				3	NUMOST	$y = 81.711 + 113.586x_1 - 109.548x_2 - 1.071x_3$	0.7298	0.5326	8.4551
				4	CMC	$y = 92.420 + 109.446x_1 - 113.752x_2 - 1.029x_3 - 46.780x_4$	0.7415	0.5498	8.3842
20	nonpathological males	right tibia	53	1	OSTHC	$y = 24.198 + 80.061x_1$	0.7386	0.5455	8.1695
				2	RATIOA	$y = 17.507 + 77.288x_1 + 43.201x_2$	0.7703	0.5934	7.8037
				3	IOSTBA	$y = 38.808 + 84.359x_1 + 47.842x_2 - 49.155x_3$	0.7945	0.6312	7.5077
				4	HCBA	$y = 52.755 + 113.896x_1 + 80.281x_2 - 77.410x_3 - 5.874x_4$	0.8187	0.6702	7.1731
21	nonpathological females	left femur	27	1	OSTHC	$y = 1.867 + 107.922x_1$	0.7703	0.5933	9.6974
				2	IOSTBA	$y = 56.419 + 119.377x_1 - 105.965x_2$	0.8272	0.6842	8.7217
				3	NUMHC	$y = 97.554 + 135.035x_1 - 144.530x_2 - 1.719x_3$	0.8772	0.7694	7.6127
				4	CDEN	$y = 38.923 + 155.491x_1 - 153.174x_2 - 2.198x_3 + 31.911x_4$	0.8918	0.7952	7.3352
				5	INDOSTA	$y = 57.731 + 142.082x_1 - 198.058x_2 - 1.848x_3 + 26.584x_4 + 624.278x_5$	0.8998	0.8097	7.2381
22	nonpathological females	right femur	36	1	OSTHC	$y = -5.096 + 115.048x_1$	0.8279	0.6854	8.1828
				2	CMC	$y = 18.500 + 94.504x_1 - 77.156x_2$	0.8635	0.7457	7.4678
23	nonpathological females	left tibia	36	1	OSTHC	$y = 20.182 + 82.333x_1$	0.7752	0.6010	9.2005
				2	CTHICK	$y = 36.241 + 76.432x_1 - 3.902x_2$	0.8388	0.7036	8.0492
				3	IOSTBA	$y = 51.959 + 83.325x_1 - 3.238x_2 - 38.220x_3$	0.8484	0.7198	7.9469
				4	OSTBA	$y = 60.843 + 102.872x_1 - 2.187x_2 - 54.720x_3 - 1.428x_4$	0.8553	0.7316	7.9027
24	nonpathological females	right tibia	37	1	OSTHC	$y = 13.904 + 90.166\,x_1$	0.8087	0.6540	8.6731
				2	IOSTBA	$y = 45.250 + 96.260x_1 - 60.748x_2$	0.8408	0.7069	8.0983
				3	INDOSTA	$y = 71.433 + 90.164x_1 - 172.232x_2 + 1280.636x_3$	0.8762	0.7677	7.3180
				4	NUMHC	$y = 112.576 + 120.351x_1 - 216.410x_2 - 948.448x_3 - 1.438x_4$	0.8917	0.7951	6.9798

TABLE 3—*Continued.*

Analysis Number	Group	Bone	*n*	Step Number	Variable Entered	Regression Equation	Multiple Correlation Coefficient	Coefficient of Determination	Standard Error of Estimate, years
				5	CTHICK	$y = 121.636 + 107.367x_1 - 199.594x_2 - 779.054x_3 - 1.268x_4 - 2.553x_5$	0.9033	0.8160	6.7199
25	upper extremities	left humerus	29	1	OSTHC	$y = -22.800 + 146.978x_1$	0.7867	0.6189	8.5181
				2	COREWT	$y = -2.221 + 135.459x_1 - 174.605x_2$	0.8784	0.7716	6.7196
				3	IHCBA	$y = 2.765 + 136.934x_1 - 181.158x_2 - 24.053x_2$	0.8874	0.7874	6.6115
				4	CDEN	$y = 48.004 + 132.876x_1 - 163.090x_2 - 22.056x_3 - 34.855x_4$	0.8975	0.8055	6.4550
				5	OSTBA	$y = 69.399 + 177.554x_1 - 138.258x_2 - 29.309x_3 - 82.086x_4 - 2.722x_5$	0.9095	0.8273	6.2135
26	upper extremities	right humerus	31	1	OSTHC	$y = -22.785 + 146.989x_1$	0.7295	0.5322	9.4605
				2	COREWT	$y = -9.854 + 145.074x_1 - 158.701x_2$	0.7883	0.6214	8.6617
				3	RATIOC	$y = -26.389 + 148.269x_1 - 145.483x_2 + 36.305x_3$	0.8087	0.6540	8.4305
				4	RATIOA	$y = -35.652 + 146.910x_1 - 139.160x_2 + 117.071x_3 - 106.455x_4$	0.8420	0.7090	7.8804
27	upper extremities	left ulna	31	1	OSTHC	$y = -6.060 + 122.880x_1$	0.6992	0.4889	10.1675
				2	IOSTBA	$y = 54.102 + 109.575x_1 - 90.145x_2$	0.7619	0.5805	9.3747
				3	NUMHC	$y = 141.925 + 192.489x_1 - 241.510x_2 - 3.122x_3$	0.8287	0.6867	8.2507
				4	CDEN	$y = 188.895 + 198.287x_1 - 237.737x_2 - 3.344x_3 - 26.201x_4$	0.8510	0.7242	7.8883
28	upper extremities	right ulna	31	1	OSTHC	$y = -1.575 + 118.104x_1$	0.6540	0.4277	10.5699
				2	OSTBA	$y = 0.550 + 194.184x_1 - 5.235x_2$	0.7251	0.5258	9.7966
				3	IOSTBA	$y = 66.925 + 230.010x_1 - 8.836x_2 - 96.633x_3$	0.7587	0.5757	9.4327
				4	COREWT	$y = 74.264 + 223.558x_1 - 8.560x_2 - 94.535x_3 - 138.297x_4$	0.7724	0.5965	9.3731

analyses performed in this study, the osteon area was selected first in all 28 cases. The *SEE* for this variable alone ranged from a high of 10.57 years in the left ulna to a low of 7.07 years in the male's right femur. After stepping had been halted, the lowest *SEE* found was 6.21 years, obtained from the analysis of the left humerus. The next lowest *SEE* was 6.41 years, obtained from the male's right femur.

Although not contributing to the reduction of the *SEE* in the stepwise linear regression analysis, analysis of age-related changes in cortical thickness and bone mineral content (g/cm and g/cm^2) revealed losses characteristic of those found in the analysis of whole bones in living U.S. whites. After age 50 males showed a 4% loss per decade for cortical thickness and a 6% loss per decade for bone mineral content (g/cm^2), while females showed an 8% loss per decade for cortical thickness and a 10% loss per decade for bone mineral content (g/cm^2).

With the regression equations generated in this study, age at death was estimated in eight forensically derived skeletons by using cores taken from femurs. The known ages at death for the eight cases ranged from 19 to 80 years. The mean known age for the forensic science series was 40.5 years and the mean estimated age was 41.5 years (Table 4). Agreement between known ages and estimated ages was good, with the greatest discrepancy found in the 80-year-old female, with a difference of five years between known and estimated ages.

Discussion

Accurately aging skeletons of persons less than 50 years old can be achieved by an experienced investigator using morphological methods. Accurately aging skeletons of persons older than 50 years requires the use of histological methods. Of the available histological aging methods, Kerley's method [*10*] has been reported to be the most accurate. However, the need for complete cross sections of bone for analysis limits access to skeletal collections, anatomical series, and forensic science cases, thus reducing widespread application of histological aging methods. Validating a method's applicability in estimating age at death in skeletons from different populations who may experience different rates of osteon turnover thus becomes difficult. Application of the same regression equations to different populations can be done only when sufficient numbers of skeletons of known age at death from each different population are analyzed. Using a small core of bone instead of a complete cross section minimizes the physical damage to a skeleton and helps ensure access to skeletons where the question of population-specific, age-related changes in osteon turnover may be addressed directly. The validity of applying these regression equations to populations other than New England whites is presently unknown but is being researched.

TABLE 4—*Eight forensic science cases of known age at death that were aged with the core technique.*

Case	Sex	Known Age, years	Estimated Age, years	Difference
1	f	19	20	+1
2	f	20	24	+4
3	f	21	19	−2
4	m	35	39	+4
5	m	39	38	−1
6	f	50	54	+4
7	m	60	63	+3
8	f	75	80	+5

From this study a *SEE* was obtained for the series that was similar to those reported by other investigators [*11,12*]. The area of cortical bone containing osteons was the single best predictor of age at death in skeletons. This finding was consistent with that reported by Ahlqvist and Damsten [*9*]. When the regression equations generated in the cadaver series were applied to the eight forensic science cases, the estimated ages corresponded well with the known ages. Other variables such as cortical thickness and bone mineral content contributed little to the reduction of the *SEE,* and this contribution was less than that of the histologically derived variables. Although these variables did not contribute to the reduction of the *SEE* in the age-estimating regression equations they do provide important information about age-related bone turnover within and between populations. In a skeletal series of known age at death patterns of age-related losses of cortical thickness and bone mineral content may provide a basis for comparisons of age-dependent bone turnover.

The results obtained from the methods of microstructure quantification used in this study are highly reproducible and are readily amenable to statistical manipulation. In skeletal series of known age at death the results obtained by these methods may be used to evaluate the size, area, and number of bone microstructures between different bones of the same individual, between sexes, between age cohorts, and between populations. Finally, the ability to obtain estimates of age at death from the analysis of bones of the upper extremities appears promising and will be the focus of additional research.

Summary

This study proposed an histological method of estimating age at death in skeletons that uses a 0.4-cm-diameter core of cortical bone. Age-estimating regression equations were generated from data obtained from the analysis of bone cores taken from femurs, tibiae, humeri, and ulnas of cadavers. When the regression equations generated in this study were applied to eight forensic science cases, accurate ages at death were estimated.

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Address requests for reprints or additional information to
D. D. Thompson, Ph.D.
Laboratory of Biological Anthropology
Department of Biobehavioral Sciences
University of Connecticut
Storrs, Conn. 06268