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Failure of Use of Cemental Annulations in Teeth to Determine the Age of Humans

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ABSTRACT: The reliability of cemental annulation counts in teeth was examined to determine its value as an age indicator for humans. One hundred extracted human teeth were embedded in polyester casting resin and sectioned using a diamond wafering saw. Thin sections were photomicrographed, projected, and counted in a double blind random method of presentation. The age of the specimen was determined by adding the number of cemental annulations counted by the observers and the eruption age of the specific tooth that was used. The data analyzed by simple regression indicated that determining chronologic age in humans from cemental annulations is not possible.

KEYWORDS: odontology, dentition, human identification, age determination

It has been recognized for over a hundred years that cementum grows continuously and forms alternating dark and light bands, or annulations, as it thickens with age [*1,2*]. Over the past 30 years scientists have reliably used cemental annulations to determine the age of various wild animals including otter [*3*], caribou [*4*], moose [*5,6*], squirrel [*7*], elk [*8*], deer [*9-13*], badger [*14*], coyote [*15*], bison [*16*], bear [*17-21*], fox [*22*], bat [*23*], and common marmoset [*24*]. Some investigators believe that cemental rings are a result of seasonal metabolic changes or nutritional variations due to hibernation; however, countable annulations have been found in nonhibernating animals as well. The underlying biological principle is not well understood.

Many investigators have recognized that cementum can be used to predict human age. Gustafson [*25*] was the first to report the use of cementum thickness along with five other features seen in teeth as a method for estimating age in humans. Recently, Azaz [*26*] and Nitzan [*27*] confirmed these observations by examining impacted teeth and reporting a direct interdependence between cementum thickness and human age.

Counting cementum annulations to predict accurately the age of primates is controversial. Wada and Hachiya [*28*] examined decalcified and undecalcified thin sections from incisors and molars of 14 Japanese monkeys. They reported that the age of these monkeys could be determined from the alternating dark and light cemental layers; however, cemental annulations in specimens of animals that had long been caged were relatively indistinct.

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Stott [24], in 1980, examined 18 marmosets and concluded that (1) all specimens demonstrated concentric dark and light cemental annulations, (2) the delineation between dentin and cementum was in some cases indistinct, and (3) interpreting annulations was sometimes difficult because of incomplete or indistinct annulations. Two years later. Stott [29] was the first investigator to correlate positively the number of cemental annulations in cross sections of undecalcified human teeth with chronological age. In contrast, Lipsinic [30] examined 35 extracted teeth, and failed to demonstrate a positive correlation between the number of incremental lines in cementum and human age.

These conflicting human studies, involving small sample sizes, have failed to elucidate the relationship of cemental annulations and age. The purpose of this study is to investigate the relationship of cemental annulations and the chronologic age of humans using a large sample size.

Materials and Methods

One hundred extracted single rooted teeth from one hundred individuals of known age (nine to seventy-eight years) were obtained.[2] The mean age of the individuals was 55.3 years with a standard deviation of 13.1 years. Collected specimens were preserved in 10% formalin. Teeth were coded to preclude any age association.

Teeth were invested by epoxy encapsulation in polyester casting resin within ½-in. (1.3-cm) butyrate plastic tubing.[3] All teeth were thin sectioned according to Stott [29] using a low speed diamond wafering saw blade.[4]

A pilot investigation was conducted to determine the type of section and thickness most conducive to visualize annulations. The entire root of ten teeth was serially sectioned (transversely and longitudinally) into 200- to 400-μm wafers. Specimens were microscopically examined. From past reports, Stott [29] and Nitzan [27], and data from the pilot investigation it was concluded that 350-μm cross sections from the midpoint of the root would produce the most representative annulations.

Finishing and polishing of specimens was necessary to eliminate grooves placed in the specimens by the diamond wafering saw blade (Fig. 1). Freshly cut specimens were rinsed with distilled water and finished with 600-grit sandpaper.[5] Each wafer was polished with 6-μm diamond compound paste on a rotary wheel.[6] All specimens were microscopically examined to determine the adequacy of finish and polish. One section per tooth from each individual was used in the study (Figs. 2 and 3).

Sections were studied and the cemental location with the most annulations was photomicrographed using black-and-white film with subdued light at ×90.[7] The photomicrographs were projected onto a screen for counting. All annulation counts were made starting from the first dark band outside of Tomes granular layer of dentin. The counting of annulations (one dark and light band constituted one annulation) was standardized among the four examiners. Viewers counted the annular structures separately in a double blind random method of presentation. The "predicted" age of each specimen was developed by taking the average of the recorded annulations counted by the four examiners and adding the mean normal tooth eruption age [31] of each specimen. The data were analyzed by computing the linear regression for each viewer and the average of the four viewers.

[2]Department of Pathology, University of Texas Health Science Center at San Antonio, TX.

[3]Polyester casting resin, Plastic Supply of S.A., 102 W. Josephine St., San Antonio, TX 78212.

[4]No. 11-1180, Isomet Low Speed Saw, Buehler, Ltd., 2120 Greenwood St., Evanston, IL 60204; and No. 11-4244 High concentration diamond wafering saw blade, Buehler, Ltd., 41 Waukegan Rd., Lake Bluff, IL 60044.

[5]Handimet Grinder with 600-grit pressure sensitive abrasive paper, Buehler, Ltd., 2120 Greenwood St., Evanston, IL 60204.

[6]No. 40-6162 Metadi diamond compound for Metallography. 6-μm paste, and No. 40-7618 Texmet 8-in. polishing cloth with adhesive backing, Buehler, Ltd., 2120 Greenwood St., Evanston, IL 60204.

[7]Panatomic X, 135-20, Eastman Kodak Company, Rochester. NY 14650.

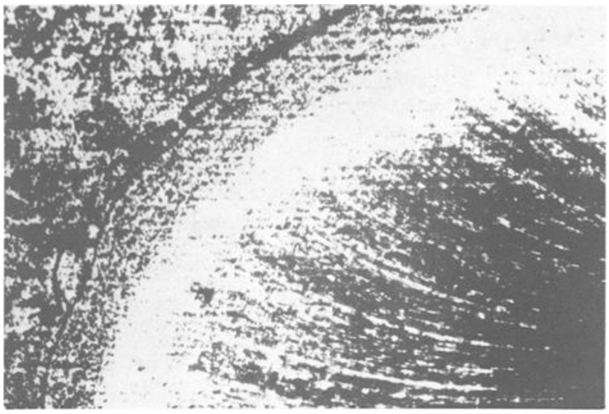

FIG. 1—*Unpolished specimen ×50.*

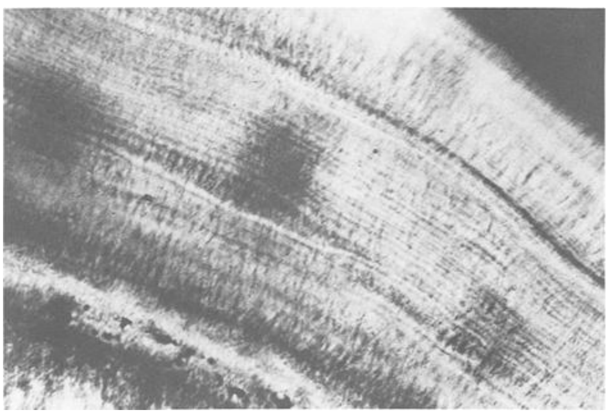

FIG. 2—*Polished specimen, 53-year-old male, ×90.*

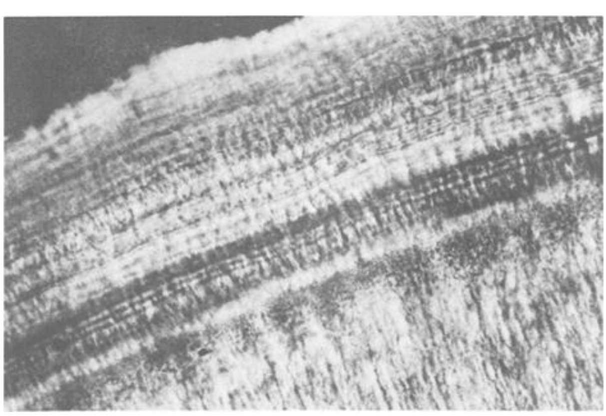

FIG. 3—*Polished specimen, 58-year-old female, ×90.*

Results

Concentric dark and light cemental bands were not clearly observed in all specimens. Twenty-nine specimens demonstrated obscured, indistinct, or no visible annulations despite finishing and polishing. These specimens were deemed inadequate for counting by one or more viewers and were eliminated from analysis. The cemental annulation counts for the remaining seventy-one specimens were used as data in analysis.

Comparison of the "predicted" age versus the chronologic age were analyzed by simple regression for each viewer as was the average value of the viewers. Only 5.7% of the specimens accurately predicted the individual's chronologic age to within five years, and over 85% of the specimens incorrectly predicted the age of the individual by greater than ten years. The slope of the regression curve for the average values was 0.2 (Fig. 4). Regression analysis revealed that while the F-ratio indicated a significant linear relationship between the estimated age and the actual age, the coefficient of determination was 0.1. Therefore, the variability of the data could not be explained by the regression model.

The regression analyses for the specimens in the less-than-35-year-age group (Fig. 5) have a higher correlation coefficient, and the estimated ages are clustered closer to the chronologic ages than the specimens from the greater-than-35-year-age group (Fig. 6).

Discussion

In recent years, many researchers have suggested the use of cementum of teeth for the determination of human chronological age. Certain characteristics of cementum suggest that it may make a reliable indicator of age. The composition of cementum is similar to bone: 60% mineral and 40% organic collagen matrix [*32*]. However, cementum is not vascularized and is resistant to resorption. Appositional growth occurs by deposition of annular structures. The thickness increases with age and varies from 20 to 200 μm.

Morphologic changes that occur in cementum are a result of function, environmental factors, and aging [*33*]. Many investigators have reported increased width of cementum in functioning and nonfunctioning teeth; however, there is no supported explanation to why humans have cemental annulations.

This study attempted to analyze the relationship of cemental annulations and human age. To minimize variables only single rooted teeth were selected for study. The midroot of each tooth was sectioned in an effort to minimize factors known to obscure annulations, that is, cementocytes, or produce variations in cementum, that is, periodontal disease and hypercementosis from local or systemic disease.

Microscopic examination of the specimens revealed only 71% of the sections had visible cemental annulations suitable for counting. The data obtained from these specimens agree with the findings of Lipsinic but disagree with the findings of Stott. The wide scattering of points (Fig. 4) indicates the great degree of variability in the cemental annulation method used to determine the age of humans. Specimens in the under-35-year-age group had a closer age correlation than the specimens in the over-35-year-age group. This correlation may be somewhat artificial because (1) a decreased variance is observed in the younger specimens as the annulation number approaches the eruption year of the tooth, (2) a smaller number of specimens were in the under-35-year-age group, and (3) possible compression and obscuration of annulations occurred in the over-35-year-age group as a result of aging.

The fact that wildlife investigators have used cemental annulations reliably to determine the age of animals conflicts with the results of this human investigation. Reasons that support the failure of this model are: humans lack seasonal variations in nutrition and metabolism, and human life expectancy is longer than the wild mammals studied to date. A better understanding of the functional relationship of cementum and aging may result from the investigation of the biochemical composition of cemental annulations.

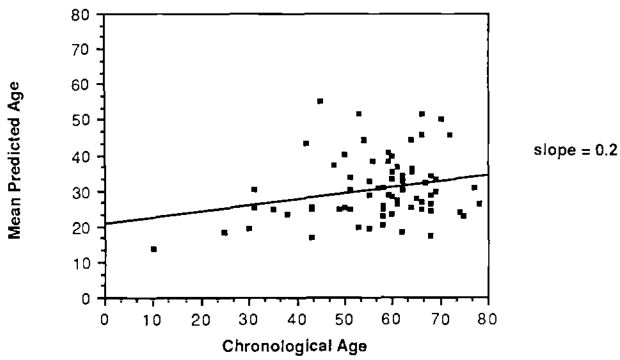

FIG. 4—*Plot of mean values of age determined by cemental annulation counts. Solid line denotes regression curve.*

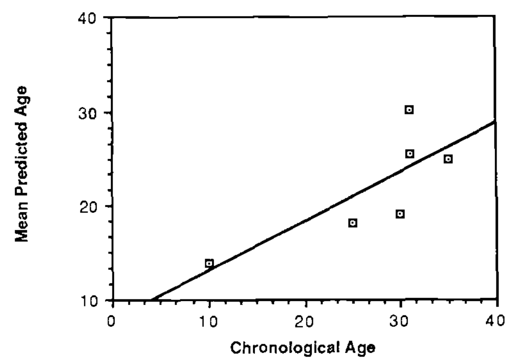

FIG. 5—*Plot of mean values of age determined by annulation counts. Solid line denotes regression curve—under-35-year-age group.*

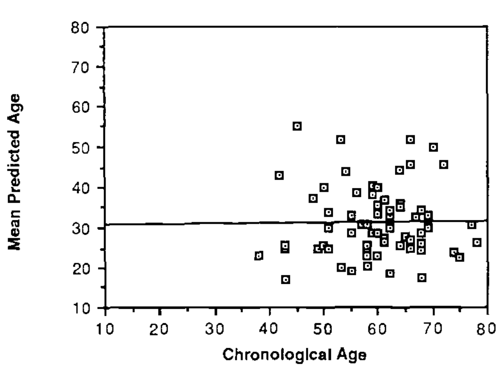

FIG. 6—*Plot of mean values of age determined by annulation counts. Solid line denotes regression curve—over-35-year-age group.*

Conclusions

Polished cross sections through the midroot of undecalcified human teeth reveal countable cemental annulations in the majority of specimens examined. Four examiners counted the annular structures in a double blind random method of presentation. The analysis of data from 71 specimens using this method indicates that determining the chronologic age of humans from cemental annulations in teeth is not possible.

Acknowledgment

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