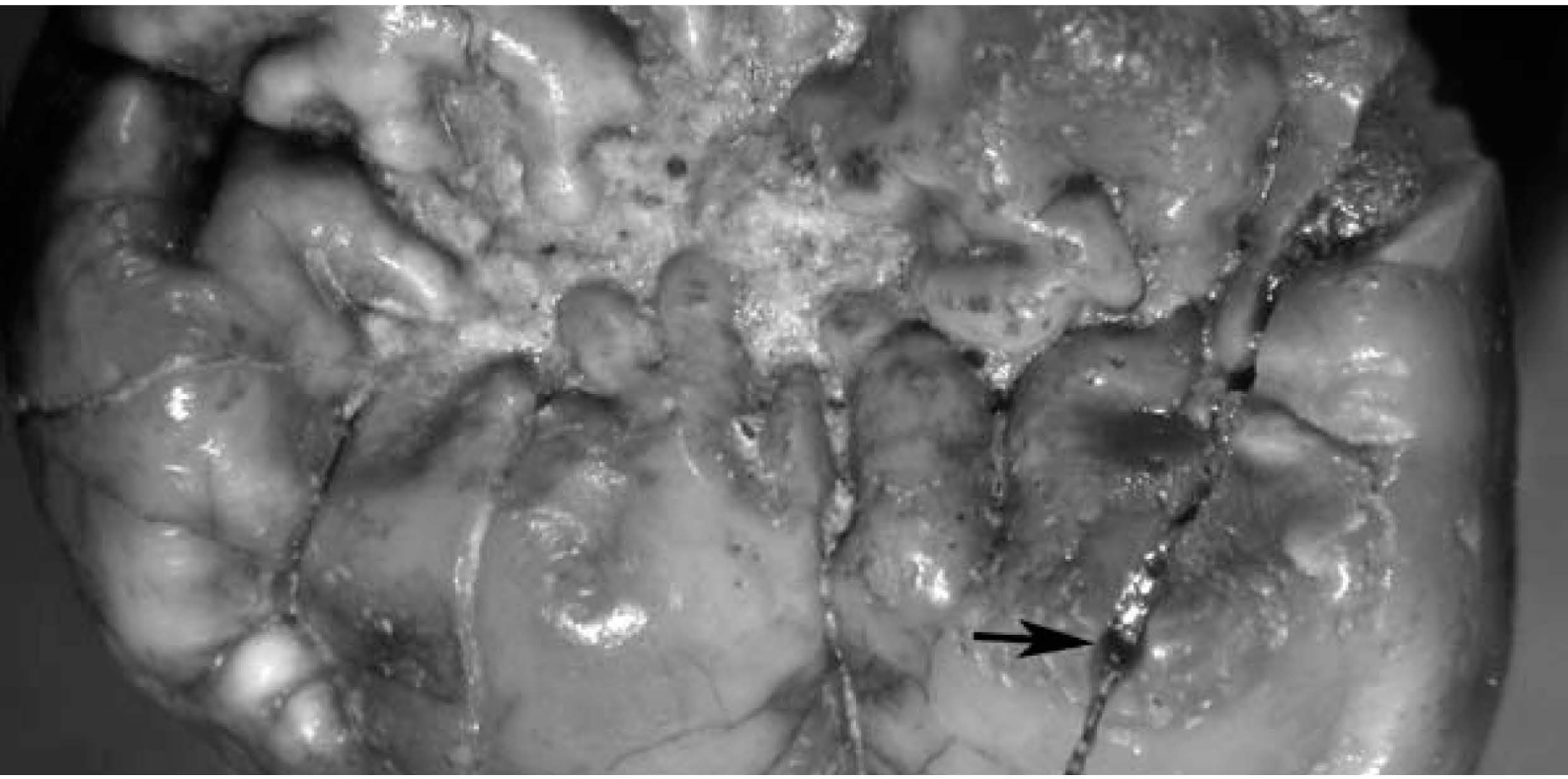

Rodrigo S. Lacruz
Fernando V. Ramirez Rozzi
Bernard A. Wood
Timothy G. Bromage

Molar development and crown areas in early *Australopithecus*

American Journal of Physical Anthropology 148: 632-640, 2012

Molar Development and Crown Areas in Early *Australopithecus*

Rodrigo S. Lacruz,[1]* Fernando V. Ramirez Rozzi,[2] Bernard A. Wood,[3,4] and Timothy G. Bromage[5,6]

[1]*Center for Craniofacial Molecular Biology, Herman Ostrow School of Dentistry, University of Southern California, Los Angeles, CA 90033*
[2]*UPR 2147 CNRS, 44 rue de l'Amiral Mouchez, Paris 75014, France*
[3]*Center for the Advanced Study of Hominid Paleobiology, George Washington University, Washington, DC 20052*
[4]*Department of Anthropology, George Washington University, Washington, DC 20052*
[5]*Department of Biomaterials and Biomimetics, New York University College of Dentistry, New York, NY 10010*
[6]*Department of Basic Science and Craniofacial Biology, New York University College of Dentistry, New York, NY 10010*

KEY WORDS *Australopithecus;* molar crown area; crown formation; Plio-Pleistocene

ABSTRACT Recent studies suggest that the hypodigms representing the two earliest *Australopithecus* (*Au. anamensis* and *Au. afarensis*) form an ancestor-descendant lineage. Understanding the details of this possible transition is important comparative evidence for assessing the likelihood of other examples of ancestor-descendant lineages within the hominin clade. To this end we have analyzed crown and cusp base areas of high resolution replicas of the mandibular molars of *Au. anamensis* (Allia Bay and Kanapoi sites) and those of *Au. afarensis* (Hadar, Laetoli, and Maka). We found no statistically significant differences in crown areas between these hypodigms although the mean of M_1 crowns was smaller in *Au. anamensis*, being the smallest of any *Australopithecus* species sampled to date. Intraspecies comparison of the areas of mesial cusps for each molar type using Wilcoxon signed rank test showed no differences for *Au. anamensis*. Significant differences were found between the protoconid and metaconid of *Au. afarensis* M_2s and M_3s. Furthermore, the area formed by the posterior cusps as a whole relative to the anterior cusps showed significant differences in *Au. afarensis* M_1s and in *Au. anamensis* M_2s but no differences were noted for M_3s of either taxon. Developmental information derived from microstructural details in enamel shows that M_1 crown formation in *Au. anamensis* is similar to *Pan* and shorter than in *H. sapiens*. Taken together, these data suggests that the overall trend in the *Au. anamensis-Au. afarensis* transition may have involved a moderate increase in M_1 crown areas with relative expansion of distal cusps. Am J Phys Anthropol 000:000–000, 2012.

The two earliest *Australopithecus* species, *Au. anamensis* and *Au. afarensis*, are widely regarded as examples of anagenetic change within a single evolving lineage (Kimbel et al., 2006; Leakey et al., 1995; Ward et al., 2001, 2010; White et al., 2006; Haile-Selassie, 2010; Haile-Selassie et al., 2010). Indeed, recent reports of *Australopithecus* from Woranso-Mille, Ethiopia suggest that any distinctions between the *Au. anamensis* and *Au. afarensis* hypodigms are so trivial that there are few morphological grounds for their being separate taxa (Haile-Selassie, 2010; Haile-Selassie et al., 2010), but if the present specific distinction is maintained then the Woranso-Mille fossils would be best ascribed to *Au. anamensis* (Haile-Selassie, 2010). Comparisons of dental characters between these hypodigms have not yet included details of molar crown and cusp base areas and only limited information is available concerning molar development (Wood et al., 1983; Beynon and Wood, 1987; Suwa et al., 1994; Dean et al., 2001; Lacruz et al., 2008; Wood, 2010). The aims of this study are to: (1) present a comparative analysis of measured crown and cusp base areas in the molars of *Au. anamensis* and *Au. Afarensis,* and (2) present data on molar development based on exposed microstructural features. These data are used to analyze aspects of molar evolution between these two hypodigms.

THE EARLIEST *AUSTRALOPITHECUS*

The chronologically oldest species of the genus *Australopithecus* is *Au. anamensis* (Leakey et al., 1995; Ward et al., 2001) (but see below). Until recently, evidence of the dentition of *Au. anamensis* was known from sediments dated to 4.2 to 3.9 Ma at Kanapoi, Allia Bay, and Asa Issie (Leakey et al., 1995; White et al., 2006). The hypodigm of the younger taxon *Au. afarensis* is largely made up of material recovered from Hadar, Laetoli, and Maka (White, 1977; Johanson et al., 1982; White et al., 2000), plus specimens recovered more recently from Dikika (Alemseged et al., 2005). Fragmentary remains have been recovered from a number of

Grant sponsor: The Leakey Foundation.

*Correspondence to: Rodrigo Lacruz, University of Southern California, Center for Craniofacial Molecular Biology, 2250 Alcazar St., CSA 103, Los Angeles, CA 90033, USA.
E-mail: rodrigo@usc.edu

Received 29 November 2011; accepted 9 April 2012

DOI 10.1002/ajpa.22089
Published online in Wiley Online Library (wileyonlinelibrary.com).

other Kenyan and Ethiopian sites, and that taxon may also have been found in Chad (e.g. Kimbel and Delezene, 2009). The temporal span of the hypodigm is between about 3.7 and 3.0 Ma (Kimbel and Delezene, 2009). There has been strong support for the proposal that *Au. anamensis* and *Au. afarensis* are related by anagenesis as an ancestral-descendant lineage (Leakey et al., 1995; Ward et al., 2001; White et al., 2006, 2009; Haile-Selassie, 2010; Haile-Selassie et al. 2010). This hypothesis has been supported by the results of numerical cladistic analysis (Strait and Grine, 2004), and analysis of dentognathic characters (Kimbel et al., 2006). More recently, this hypothesis has been bolstered by the discovery of hominin fossils at Woranso-Mille in Ethiopia that date to between 3.57 and 3.8 Ma i.e., to the temporal interval between the hypodigms of *Au. anamensis* and *Au. afarensis* (Haile-Selassie, 2010; Haile-Selassie et al., 2010). These remains preserve a suite of morphological characters some of which are intermediate between *Au. anamensis* and *Au. afarensis* and some of which are shared by one or both of those taxa (Haile-Selassie, 2010; Haile-Selassie et al., 2010). Based on the Woranso-Mille evidence, distinctions between *Au. anamensis* and *Au. afarensis* were deemed "confusing and unwarranted" (Haile-Selassie, 2010). However, for the purpose of this study, we have maintained the taxonomic separation of these two hypodigms.

Crown areas and enamel development

The overall crown areas and the areas of the individual cusp boundaries of mandibular molars have been used to assess patterns of morphological variation within Plio-Pleistocene hominin species (e.g., Wood et al., 1983; Suwa et al., 1994). The methods used in these studies have been adapted for the analysis of the crown morphology of premolars (Wood et. al., 1983; Suwa, 1988) and maxillary molars (Bailey, 2004; Moggi-Cecchi and Boccone, 2007; Grine et al., 2009; Quam et al., 2009). In addition to the crown base area data, this study also includes evidence about enamel microstructure. The enamel microstructure of primates and that of many other mammalian taxa preserves daily (or short period) growth markings called cross-striations, which represent a day's worth of matrix secretion by the enamel forming cells, or ameloblasts. Besides cross-striations, enamel preserves approximately circaseptan long period growth markings called striae of Retzius, which may form perikymata where they reach the lateral enamel surface (Boyde, 1989). The number of cross striations between adjacent striae is known as the periodicity, or as the striae repeat interval and the distance between the cross-striations corresponds to the amount of enamel added to the thickness of the crown in a day, also known as the daily secretion rate. Further details on these markings can be found in Boyde (1989).

MATERIALS

Data were collected from the originals of the teeth of *Au. anamensis* reported in Leakey et al. (1995) and Ward et al. (2001). The *Au. anamensis* samples from Asa Issie and Woranso-Mille were not included in this analysis. A total of 17 molars of *Au. anamensis* teeth were analyzed (Appendix). The sample consists of M1 ($n = 7$); M2 ($n = 6$) and M3 ($n = 4$). Because our cast collection of *Au. afarensis* molars was incomplete, we obtained a more complete data set kindly provided by Dr. Gen Suwa (referred to hereafter as the GS data set) which was originally reported in Suwa et al., (1994) (Appendix). In addition to these data, the Maka sample (White et al., 2000) was included, and for consistency, we used values for the Maka sample measured by GS. A total of 48 teeth of *Au. afarensis* were analyzed, which excludes the Dikika sample that was not examined for this study.

METHODS

There are subtle differences in the methods used to measure cusp and cusp boundaries in molars (Wood et al., 1983; Suwa et al., 1994; Bailey et al., 2004). The main differences are in the orientation or placing of the specimen so as to maximize the occlusal crown area. Whereas Wood et al. (1983) used the cervical line as a plane of reference; others have used the occlusal fovea to maximize the occlusal crown area (Suwa et al., 1994, 1996). Despite these differences in the method, studies suggest that it has no significant impact on the resulting measurements (Suwa et al., 1994; Bailey et al., 2004; Grine et al., 2009). In our study we followed the protocol described in Suwa et al., (1994).

For *Au. afarensis* we used the GS data set on cusp and crown areas. For *Au. anamensis*, cusp areas were measured from images taken on high resolution casts of the specimens made by us using Coltène President in its "light and putty" variant. Casts were photographed using a Zeiss stereo microscope coupled with Nikon Coolpix 4500 at 6× magnification. Total crown base areas (CBA) as well as individual cusp areas were measured using ImageJ software. Individual cusp base areas were measured following Wood et al. (1983) in which surface areas of accessory cusps were divided equally and added to the adjacent principal cusps. Crowns in which wear had removed substantial lengths of the primary or secondary fissures or specimens that required extensive reconstruction of the crown outline were not included in this analysis. Each specimen was measured twice and the mean of the two measurements was taken to be the value for that specimen. If antimeres were available, the mean of both areas was used. To assess potential differences between *Au. anamensis* and *Au. afarensis* molars using the GS data set for *Au. afarensis*, we analyzed interobserver error differences by comparing the same *Au. afarensis* molars measured by us with measurements of the same specimens made by GS. Two tailed Student's *t*-test was used to assess these differences.

Univariate analysis of cusp and crown areas includes basic descriptive statistics as well as the nonparametric Mann-Whitney *U* test for comparing the cusp and crown base areas between *Au. anamensis* and *Au. afarensis*. We compared not only the total crown base area (CBA) of the molars but also the absolute sizes of the individual cusps and their relative contribution to the composition of the occlusal surface of the whole crown. To compare differences in cusp size within the molar series of each taxon (i.e., to compare the protoconid and metaconid of each tooth type in each species), or to compare the combined areas of the distal cusps relative to anterior cusps, we employed the nonparametric Wilcoxon signed rank test.

Molar development

Materials. Two naturally fractured *Au. anamensis* molars, the M_1 KNM-KP 31712j and a possible M_1

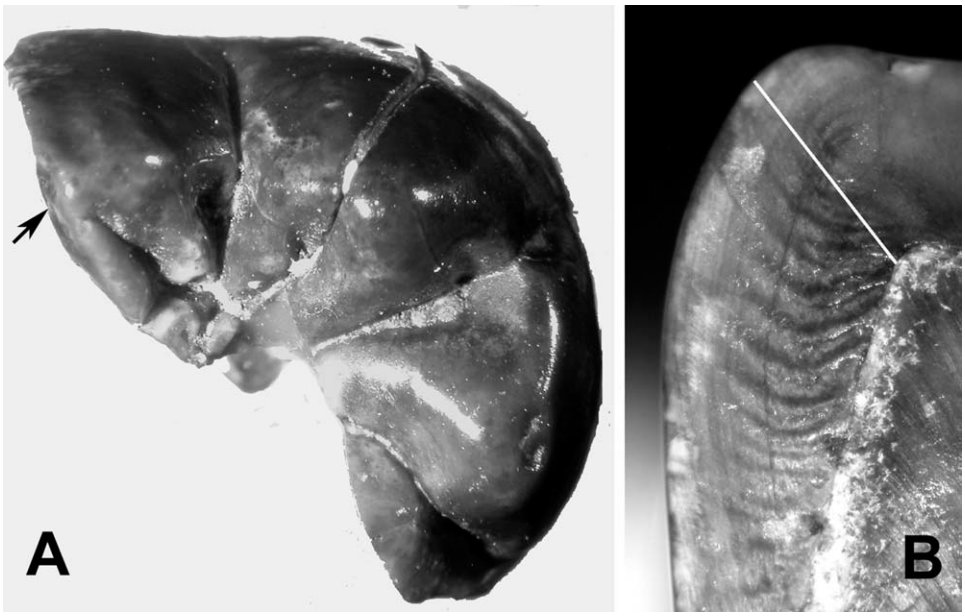

Fig. 1. **A**: The occlusal view of KNM-ER 30749, an incomplete left M_1 (?) crown showing a fracture on the metaconid cusp tip (arrowed). **B**: Lateral view of the metaconid indicating by a white line the approximate area where we measured the cuspal enamel thickness. Note the concave shape of the Hunter Schreger bands (HSB) in (B) in this specimen from the younger *Au. anamensis* site of Allia Bay, which contrasts with the shape of HSB described in some *Au. afarensis* molars (Lacruz and Ramirez Rozzi, 2010).

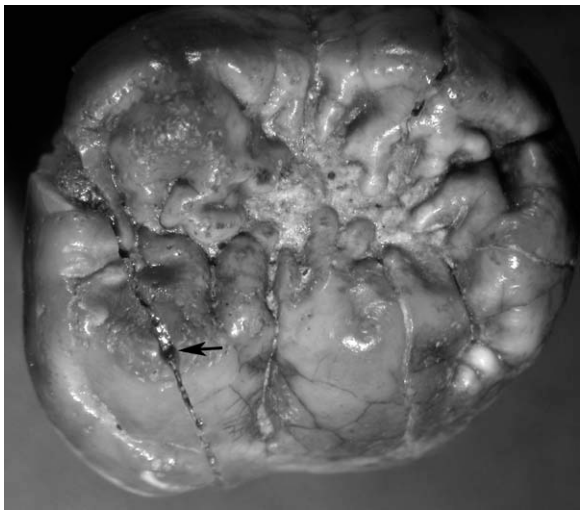

Fig. 2. Occlusal view of KNM-KP 31712j, an M_1 of *Au. anamensis*. The protoconid cusp (arrowed) is perfectly fractured naturally almost at the tip as seen in the image. We were able to unglue this specimen to study its microstructure.

(KNM-ER 30749; Ward et al., 2001) were used to estimate cusp and crown formation time. These specimens were selected because they showed natural fractures that passed near the dentine horn broadly oriented in the buccolingual plane (Figs. 1A,B and 2). For detailed descriptions of each specimen see Ward et al., (2001; p. 293 [KNM-KP 31712j] and p. 325 [KNM-ER 30749]). This latter specimen had been previously sectioned and cusp formation time for the hypoconid reported in Ward et al. (2001). Here we report on the metaconid cusp formation of this molar and on the protoconid of KNM-KP31712j.

Methods. We have previously described in detail the protocol for estimating crown formation time in hominin molars (Lacruz et al., 2006; Lacruz and Ramirez Rozzi, 2010). Following the method of Reid et al., (1998) the lateral perikymata or striae of Retzius are counted and the total number is multiplied by the periodicity, which provides the lateral formation time of the crown. For the occlusal or cuspal enamel we used the method originally described in Beynon et al. (1991). The linear enamel thickness is measured from a point just below the gnarled enamel at the EDJ and the region in the outer enamel where the last lateral stria is identified. This measurement is then divided by the average value of the cross striation length, providing the cuspal or occlusal formation time. The average values of cuspal cross striations for *Au. anamensis* and *Au. afarensis* were reported in Lacruz et al. (2008). Crown formation time is the sum of lateral and cuspal enamel.

RESULTS

Interobserver error

Table 1 shows descriptive statistics for the *Au. afarensis* molars measured by GS and by us comparing both data sets. Two-tailed Student's *t*-test reveals no significant differences between measurements of the same samples made by different observers using a similar method. In fact, our results and those obtained by GS are strikingly similar when the overall mean values are considered. However, differences in individual cusp measurements differed by as much as 25% in some cases, although most commonly the differences were around 10–15%.

Crown and cusp base areas

Table 2 shows descriptive statistics of individual cusps and crown base areas of lower molars of *Au. anamensis*

TABLE 1. Interobserver error measurements were assessed using t-test comparison of the means of the cusp and crown areas for Au. afarensis molars measured by us and those measured in Suwa et al. (1994)

	Protoconid	Metaconid	Hypoconid	Entoconid	Hypoconulid	Total area
M1 this study	34.3 (5.8) $n = 6$	30.27 (5.9) $n = 6$	32.1 (3.7) $n = 6$	21.44 (4.1) $n = 6$	21.6 (4.1) $n = 6$	139.12 (15.8) $n = 8$
M1 GS	32.4 (3.7) $n = 6$	30.3 (4.47) $n = 6$	29.8 (2.5) $n = 6$	24.84 (5.5) $n = 6$	22.1 (2.8) $n = 6$	138.25 (14.7) $n = 8$
P value	$P = 0.51$; $n = 6$	$P = 0.99$; $n = 6$	$P = 0.23$; $n = 6$	$P = 0.25$; $n = 6$	$P = 0.79$; $n = 6$	$P = 0.91$; $n = 8$
M2 this study	44.96 (7.4) $n = 6$	36.79 (8.2) $n = 6$	32.96 (5.7) $n = 6$	22.31 (2.9) $n = 6$	27.13 (6.1) $n = 6$	163.34 (20.2) $n = 8$
M2 GS	45.34 (7.1) $n = 6$	38.18 (7.9) $n = 6$	32. 14 (4.0) $n = 6$	24.64 (1.8) $n = 6$	25.75 (6.5) $n = 6$	166.10 (19.85) $n = 8$
P value	$P = 0.92$; $n = 6$	$P = 0.77$; $n = 6$	$P = 0.95$; $n = 6$	$P = 0.13$; $n = 6$	$P = 0.71$; $n = 6$	$P = 0.78$; $n = 8$
M3 this study	44.76 (4.0) $n = 5$	42.57 (2.4) $n = 5$	27.90 (4.3) $n = 5$	21.75 (5.8) $n = 5$	33.55 (6.6) $n = 5$	168.51 (16.1) $n = 7$
M3 GS	45.71 (6.5) $n = 5$	41.01 (4.3) $n = 5$	27. 15 (3.6) $n = 5$	28.97 (5.1) $n = 5$	30.56 (7.2) $n = 5$	169.66 (17.1) $n = 7$
P value	$P = 0.79$; $n = 5$	$P = 0.50$; $n = 5$	$P = 0.77$; $n = 5$	$P = 0.07$; $n = 5$	$P = 0.51$; $n = 5$	$P = 0.99$; $n = 7$

and *Au. afarensis*. *Au. anamensis* M_1 crown base areas are somewhat smaller than those of *Au. afarensis* showing smaller individual cusp means, which in the case of entoconid and hypoconulid, were significantly different ($P < 0.05$). Averages for M_2s crowns were slightly higher in *Au. anamensis* whereas M_3s were moderately smaller. Significant differences were observed for M_2 hypoconulids and for M_3 entoconids (Table 2). A graphical representation (box plots) of the total crown base areas of molars is shown in Figure 3. The Wilcoxon signed rank test was used to compare the absolute size of the principal mesial cusps of each molar type between the hypodigms (Table 3). For *Au. anamensis*, no statistical differences between the protoconid and metaconid were identified for any of the molar types. For *Au. afarensis*, no differences were identified in M_1s, whereas M_2s and M_3s showed statistical differences between the main cusps. Despite absolute differences between the areas of the protoconid and metaconid in *Au. afarensis* M_2s and M_3s, Table 4 shows that there were no major changes in the contribution of individual cusp base areas to the total CBA between the two *Australopithecus* hypodigms. Table 3 also shows results of Wilcoxon signed rank test comparing the area formed by the combined measurements of the posterior cusps relative to the areas formed by the anterior cusps. For M_1s, the increase in the areas formed by the posterior cusps was significant in *Au. afarensis*, but for M_2s, an increase in the areas formed by the posterior cusps was significant in *Au. anamensis*. No differences were identified in M_3s in either hypodigm.

Molar development

Using the method described in Lacruz et al. (2008) we were able to obtain a periodicity of 7 days for the *Au. anamensis* specimen KNM-KP 31712j, which together with the periodicities for two other *Au. anamensis* reported in Ward et al. (2001), brings to a total of three the number of *Au. anamensis* teeth with a known periodicity; in all three cases it is 7 days. Thus, we use the 7 day periodicity to estimate the cuspal and crown formation times of *Au. anamensis* M_1s (Table 5); the former were calculated for the metaconid (KNM-ER 30749) and protoconid (KNM-KP 31712j). We used the average of the daily secretion rates from Lacruz et al. (2008) (i.e., a mean occlusal DSR of 4.7 μm). KNM-ER 30749 was slightly worn and we estimated 200 μm of missing cuspal enamel should be added to our estimations of linear enamel thickness. KNM-KP 31712j is described as an unworn M_1 crown (Ward et al., 2001, p. 293), but in this case we added 100 μm as the fracture plane appears to have missed the very tip of the cusp (Fig. 2) (Table 5). Cusp formation times for each cusp are 2.2 years for the metaconid of KNM-ER 30749 and 2.1 years for the protoconid of KNM-KP 31712j.

DISCUSSION

Prior to this study, only qualitative descriptions of cusp areas of *Au. anamensis* were available (Ward et al., 2001). For *Au. afarensis*, cusp and crown base areas were quantified and reported in Suwa et al., (1994) and Bromage et al., (1995). Our analysis of *Au. afarensis* molars presented here differs from previous studies in that we have included the Maka dental sample that was reported after the publication of Suwa et al.'s and Bromage et al.'s analyses.

Interobserver differences between our analysis and that reported by Suwa et al., (1994) using the same specimens reveal very minor differences in results. Although minor differences were noted between the means largely ranging from 10 to 15%, no statistically significant differences were identified (Table 1). This suggests that we can confidently use the GS data on *Au. afarensis* to compare it with our data on *Au. anamensis* molars.

Differences in crown base areas between *Au. anamensis* and *Au. afarensis* largely concern the smaller M_1s of *Au. anamensis* ($P = 0.073$) (Table 2). One of the *Au. anamensis* M_1 specimens (KNM-KP 30500) was markedly larger than the remaining M_1 of this group available for study and was larger than all the known *Au. afarensis* M_1s. It is also noticeable that the crown of the M_2 of KNM-KP 30500 is also larger than all of the *Au. afarensis* M_2s. Box plots of crown areas are shown in Figure 3.

Comparing the average values of the *Au. anamensis* crown base areas with the mean values for *Au. africanus* in Suwa et al., (1994) and Bromage et al., (1995), *Au. anamensis* crown areas are smaller than the *Au. africanus* mean for each molar type (Fig. 4). Compared with the East and southern African *Paranthropus* samples, *Au. anamensis* crown base area values are smaller for all tooth types. When compared with the mean of the values of the early *Homo* group reported by Suwa et al., (1994), *Au. anamensis* has moderately larger M_2 and similar M_3 mean crown base areas, whereas the M_1s are smaller in the *Au. anamensis* hypodigm. The picture that emerges from these comparisons is that the *Au. anamensis* hypodigm has the smallest M_1 crown areas among the early *Australopithecus* samples analyzed to date, a finding consistent with metrical data on standard MD-BL measurements (Suwa et al., 2009; Kimbel and Delezene, 2009). Although some variation on a specimen by specimen basis is noted, Figure 4 also indicates that the overall trend in crown base area size is $M_1 < M_2 >$

TABLE 2. Mean, range, and standard deviation for cusp and total crown areas of lower molars of Au. anamensis and Au. afarensis

		Protoconid	Metaconid	Hypoconid	Entoconid	Hypoconulid	Total crown
Au. anamensis	M1	27.5 (22.1–35.9) (5.8)	27.2 (23.9–33.2) (3.6)	23.5 (18.5–32.3) (5.9)	19.1 (15.5–21.4) (2.4)	17.1 (11.9–21.5) (3.8)	124.5 (99.8–166.2) (24.5)
		N = 5	*N* = 5	*N* = 5	*N* = 5	*N* = 5	*N* = 7
Au. afarensis	M1	32.5 (25.7–36.1) (3.6)	30.9 (22.6–35.6) (4.4)	30.2 (25.4–33.1) (2.4)	24.6 (19.6–33.2) (4.9)	22.1 (18.6–26.9) (2.8)	141.1 (111.9–160.7) (15.4)
		N = 7	*N* = 7	*N* = 8	*N* = 8	*N* = 8	*N* = 13
P values		*P* = 0.123	*P* = 0.167	*P* = 0.079	*P* = 0.040	*P* = 0.028	*P* = 0.073*
Au. anamensis	M2	43.2 (34.6–51.4) (7.6)	39.8 (35.8–47.2) (4.3)	34.4 (28.4–42.5) (5.2)	25.0 (18.1–29.8) (4.2)	29.0 (27.6–31.0) (1.4)	170.7 (145.7–202.3) (18.7)
		N = 5	*N* = 5	*N* = 5	*N* = 5	*N* = 5	*N* = 6
Au. afarensis	M2	42.48 (32.0–57.2) (7.4)	36.3 (25.5–44.7) (7.5)	31.8 (25.5–39.0) (4.4)	25.8 (21.2–35.3) (4.0)	24.8 (19.2–36.5) (4.9)	162.4 (129.6–197.6) (20.9)
		N = 10	*N* = 10	*N* = 12	*N* = 12	*N* = 12	*N* = 20
P values		*P* = 0.806	*P* = 0.540	*P* = 0.343	*P* = 0.598	*P* = 0.035	*P* = 0.394
Au. anamensis	M3	42.7 (40.7–44.5) (1.9)	33.9 (32.3–36.2) (2.1)	26.5 (22.0–30.3) (4.2)	19.2 (16.4–22.2) (2.9)	33.7 (24.6–39.0) (7.7)	161.1 (147.8–177.1) (12.8)
		N = 3	*N* = 3	*N* = 3	*N* = 3	*N* = 3	*N* = 4
Au. afarensis	M3	42.5 (34.0–56.2) (6.5)	36.4 (26.7–43.5) (5.4)	28.0 (21.5–34.3) (4.1)	27.3 (22.0–36.3) (4.1)	28.6 (21.5–38.7) (5.4)	163.6 (140.8–202.9) (15.9)
		N = 10	*N* = 10	*N* = 11	*N* = 11	*N* = 11	*N* = 15
P values		*P* = 0.996	*P* = 0.398	*P* = 0.586	*P* = 0.016	*P* = 0.312	*P* = 0.073

P values indicate results of Mann-Whitney U test comparing values of each cusp between hypodigms.

M_3 in *Au. anamensis* and *Au. africanus*. This is a similar trend to that described for early *Homo* (Suwa et al., 1994). For the *Au. afarensis* hypodigm, M_2 crown base area is moderately smaller than M_3 (Fig. 4), as was also observed in *P. boisei* (Wood et al., 1983; Suwa et al., 1994).

Molar crown development

Information on the timing of the development of *Au. anamensis* molars was provided in Ward et al. (2001). The cusp formation times published in Ward et al. (2001) are as follows: KNM-KP 30748 protocone = 2.28 years and KNM-ER 30749 hypoconid = 2.72 years. Ward et al. (2001) indicated that "*the greater time for cusp formation in the KNM-ER 30749 hypoconid than the KNM-KP 30748 protocone suggests that this specimen belongs more distal in the tooth row than does KNM-KP 30748.*" Indeed M_1s have shorter crown formation than M_2s and M_3s in humans and in chimpanzees (Reid and Dean, 2006; Smith et al., 2007). However, in Ward et al., (2001; p. 325) it is indicated that KNM-ER 30749 is a left M_1. Given the uncertainty concerning KNM-ER 30749 and because of a method recently reported to assess crown formation time based on cusp formation time (see below), it is perhaps more informative to focus our attention on the protoconid of KNM-KP 31712j (M_1) to assess crown formation. The cusp formation time obtained was 2.1 years, which is in line with the values reported by Ward et al., (2001) for a maxillary protocone.

We have previously estimated molar crown formation times in molars of *Au. afarensis*, *Au. Africanus,* and *P. robustus* from cusp formation times using a method originally described by Ramirez Rozzi (1993) in an analysis of Ethiopian specimens from the Omo Formation (Lacruz et al., 2006; Lacruz and Ramirez Rozzi, 2010). In this study we use a different method. The reason to do so is the difficulty in following the last cervical stria to its corresponding perikyma and following this to the distal moiety of the molar, as we had previously done (Ramirez Rozzi, 1993; Lacruz et al., 2006). Thus we relied on the results of a recent study of modern human M_1 crown development that showed that the time taken to develop the protoconid approximates (with a small, c. 7%, margin of error) to the time taken to form the whole crown (Mahoney, 2008). Using this relationship and the estimate of the time to form the protoconid of KNM-KP 31712j (2.1 years or 759 days), we can obtain an estimate of 2.2 years (759 days + 7% or 53 days = 812 days) for the formation time of the entire crown. We did not use the same method to assess crown formation time in KNM-ER 30749 because inferring total crown formation time based on metaconid formation times had a noticeably greater error (Mahoney, 2008). However, the crown formation time for this molar based on our results and those reported in Ward et al. (2001) can be estimated to be between 2.2 and 2.7 years. The estimated crown formation time of 2.2 years for KNM-KP 31712j (M_1) is similar to the reported average value of chimpanzee (*Pan troglodytes*) M_1 crown formation (Smith et al., 2007) suggesting that this may be the symplesiomorpic condition for early hominins. The crown formation time is also shorter than the corresponding values for *Homo sapiens* (Reid and Dean, 2006). A cautionary note should be included here. We had originally reported on the meta-

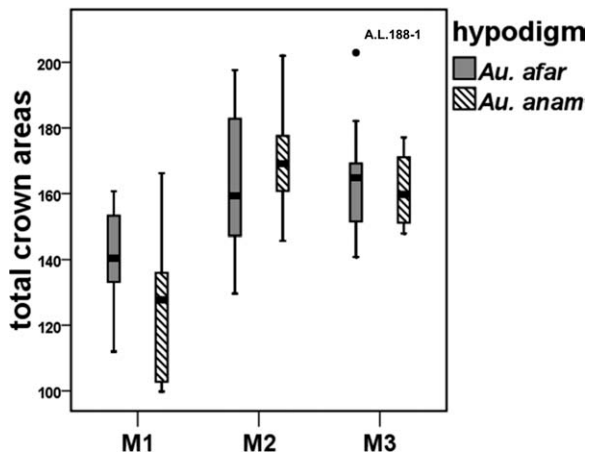

Fig. 3. Box plots of total crown areas for each molar type in lower molars of *Au. anamensis* and *Au. afarensis*.

conid cusp formation time of A.L. 333-52 which was described by Johanson et al. (1982) as an M_1. However, it has come to our attention that this tooth may be best classified as an M_2 and that the cusp analyzed by us was not a metaconid but an entoconid (Gen Suwa pers. comm.). Therefore, data obtained here for *Au. anamensis* M_1s may not be directly compared with like-tooth type in *Au. afarensis*.

It is well known that the timing of M_1 eruption is highly correlated with life history-related variables (i.e., cranial capacity) (Smith, 1991). For hominins, however, data on molar eruption times are scarce (e.g., Dean, 2010) but in the absence of such information, M_1 crown formation time could possibly be used as a broad indicator of hominin life history, although this issue remains a subject of study (Macho, 2001; Kelley and Smith, 2003; Schwartz et al., 2005). In hominins, this approach is hampered because of the few data that are available on M_1 crown formation time (Lacruz and Ramirez Rozzi, 2010). Despite these uncertainties, M_1 crown formation time can be used to predict the age at death of certain individuals (e.g. Lacruz et al., 2005). Using our reported M_1 crown formation for *Au. anamensis* (2.2 years, Table 5), together with the average rate of root growth in *Au. anamensis*, apes or in modern humans of between 1 and 2 mm of the first root formed (c. 6.0 microns/day) (Dean, 2010), this information may be used to estimate age at death of juvenile *Au. anamensis* fossils in the future. Furthermore, because the time invested by each species in forming a tooth depends on the schedule of growth and development of that species and the time available to do this (e.g. Bromage, 1987; Dean, 2006; Lacruz et al., 2008) the data on crown formation time presented here and the reported data on *Au. anamensis* root formation (Dean, 2010) suggests potentially similar developmental schedules for *Au. anamensis* and *Pan*. This matches what has been proposed some time ago for *Au. afarensis* (Bromage and Dean, 1985; Anemone, 2002).

TABLE 3. Top: Intraspecies comparison of the areas of the main anterior cusps (protoconid vs. metaconid) for each molar type in Au. anamensis and Au. afarensis lower molars using Wilcoxon signed rank test; Bottom: Comparison of the relative areas of the individual posterior cusps combined with the combined areas of the anterior cusps for each molar type using Wilcoxon signed rank test

	Protoconid vs. metaconid		
Hypodigm	M1	M2	M3
Au. anamensis	$P = 0.502$	$P = 0.225$	$P = 0.109$
Au. afarensis	$P = 0.176$	$P = 0.005$	$P = 0.022$
	Anterior vs. posterior cusps		
	M1	M2	M3
Au. anamensis	$P = 0.08$	$P = 0.043$	$P = 0.109$
Au. afarensis	$P = 0.018$	$P = 0.169$	$P = 0.114$

Morphological changes in *Australopithecus* molars

Among the differences identified between the *Au. anamensis* and *Au. afarensis* fossils, including the Woranso-Mille material, the mandibular molars of *Au. anamensis* are described as being lower crowned than those of *Au. afarensis* and possessing "sloping buccal sides" (Leakey et al., 1995). The maxillary molars possess "trigons much wider than talons" (Leakey et al., 1995). However, the size of the permanent postcanine teeth was considered to be similar in the two species (Haile-Selassie et al., 2010; Ward et al., 2001). The occlusal morphology of mandibular molars (i.e., as judged by the observed size of main cusps) was also described as being similar in these taxa although no quantitative data was reported (Ward et al., 2001). Descriptions of the occlusal areas of lower molars indicated that the largest cusps in *Au. anamensis* were the protoconid and metaconid (Ward et al., 2001, p.350). It was also noted that the fissure patterns of the *Au. anamensis* molars exhibited enough variation to suggest caution about the use of such features for taxonomic purposes (Ward et al., 2001, p. 350).

The results of the quantitative analyses presented here, which do not include the Woranso-Mille or Asa Issie *Au. anamensis*, show that the crown base area values of *Au. anamensis* M_1s are smaller than those for *Au. afarensis*, whereas M_2 are slightly bigger and have similar M_3 crown. Few statistically significant differences were identified for any of the crowns of the molar types although differences were identified in some of the distal cusps (entoconid and hypoconulid) of all molar positions. These differences may be related to the complexities of the distal moiety of lower molars which makes difficult to clearly identify the boundaries of the distal cusps

TABLE 4. Percentage that each individual cusp area contributes to the total crown area in lower molars of Au. anamensis and Au. afarensis

Hypodigm	Molar	Protoconid	Metaconid	Hypoconid	Entoconid	Hypoconulid
Au. anamensis	M1	23.6% (sd 2.3)	23.3% (sd 1.9)	20.6% (sd 2.1)	16.7% (sd 3.0)	15.7% (sd 2.4)
Au. afarensis	M1	23.4 % (sd 1.4)	22.0% (sd 0.9)	21.5% (sd 0.9)	17.4% (sd 1.9)	15.7% (sd 1.1)
Au. anamensis	M2	24.9% (sd 2.7)	23.1% (sd 0.9)	19.5% (sd 1.1)	15.6% (sd 1.5)	16.7% (sd 1.6)
Au. afarensis	M2	26.1% (sd 1.9)	22.9% (sd .18)	19.7% (sd 1.5)	16.1% (sd 2.1)	15.2% (sd 1.9)
Au. anamensis	M3	26.2% (sd 2.4)	22.5% (sd 2.0)	16.4% (sd 2.0)	13.4% (sd 4.1)	21.4% (sd 3.8)
Au. afarensis	M3	26.1% (sd 1.6)	22.8% (sd 2.7)	17.0% (sd 1.7)	17.1% (sd 1.6)	17.0% (sd 2.6)

TABLE 5. Crown formation time in Au. anamensis molars

Locality	Allia Bay	Kanapoi
Specimen	KNM-ER 30749	KNM-KP 31712j
Cusp	Metaconid	Protoconid
LET	1,147 μm	1,100 μm
C/LET	1,347 μm	1,200 μm
Lateral/SR	73	72
Periodicity	7	7
AV Oc/DSR	(4.7 μm)	(4.7 μm)
LFT/d	511	504
OFT/d	286	255
Cusp FT/years	2.2	2.1
Total CFT/years	?	2.2

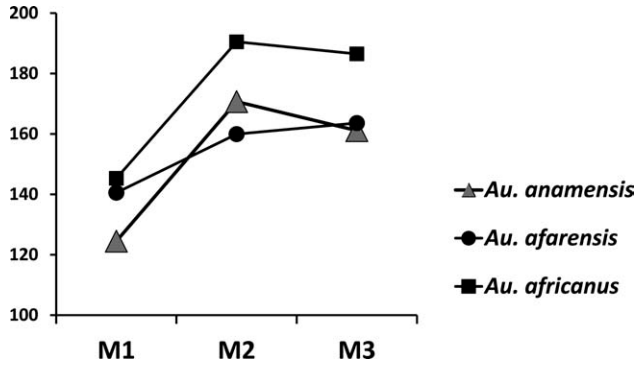

Fig. 4. Comparison of the averages of crown areas in *Australopithecus* molars. The molars of *Au. anamensis* are smaller than other *Australopithecus.* These values are, however similar, to those reported for early *Homo* (Suwa et al., 1994). It is also noticeable that there has been an increase in M_1 crown areas from the oldest *Australopithecus* (*Au. anamensis*) to the youngest species of the genus (*Au. africanus*). *Au. africanus* data were obtained from Suwa et al. (1994).

(Suwa et al. 1994, 1996; Ward et al., 2001; Bailey et al., 2004). Although Wilcoxon signed rank test showed no statistically significant differences between the protoconid and metaconid in the *Au. anamensis* molar series, we identified significant differences for *Au. afarensis* M_2s and M_3s (Table 3). Furthermore, when the contribution of the individual cusps areas is considered in relation to the whole crown for all molar types the results for the two taxa were similar (Table 4). Table 2 also indicates that for the M_2s and M_3s of *Au. anamensis* and *Au. afarensis*, the protoconid>metaconid>hypoconid, whereas in the M_1s the mesial cusps are sub-equal in size. Finally, the expansion of the distal moiety of *Au. afarensis* M_1s was significant relative to the area formed by the anterior cusps. For M_2s this is noted in the *Au. anamensis* hypodigm only, whereas no differences were noted in M_3s for either hypodigm. These data indicate that for the lower molars, the transition from *Au. anamensis* to *Au. afarensis* may have included a moderate increase in M_1 crown area with expansion of the distal moiety of this tooth.

CONCLUSION

Our analysis suggests that there may have been a moderate increase in M_1 crown base area from *Au. anamensis* to *Au. afarensis*. Previous commentators have suggested that changes in tooth crown size, which we here extend to include changes in overall crown and individual cusp areas, are indicative of changes in the properties of foods consumed (Lucas et al., 1986). In keeping with this notion and with previous studies (Teaford and Ungar, 2000; Ward et al., 2001, 2010; White et al., 2006), a modest increase in M_1 crown base area from *Au. anamensis* to *Au. afarensis* is consistent with a modest shift in dietary adaptations between these two groups (but see Grine et al., 2006; Ungar et al., 2010). This adaptive shift appears to be supported by additional material of *Au. anamensis* that is not yet fully described (Ward et al., 2010).

This study contributes to a better understanding of the evolution of mandibular molars within *Australopithecus* and provides additional evidence about the nature of the relationships between *Au. anamensis* and to *Au. afarensis.*

ACKNOWLEDGMENT

The authors express their gratitude to the Kenyan and Ethiopian Governments, the National Museums of Kenya and the staff at the Ethiopian National Museum, Addis Ababa, and to Mamitu Yilma and Emma Mbua for access to the fossil material in their care. They are indebted to Meave Leakey for access to the *Au. anamensis* material. Gen Suwa is thanked for allowing them to use his *Au. afarensis* data and for critically reviewing previous drafts which substantially improved this manuscript. They also thank an anonymous reviewer for helpful comments.

LITERATURE CITED

Anemone RL. 2002. Dental development and life history in hominid evolution. In: Minugh-Purvis N, McNamara KJ, editors. Human evolution through developmental change. Baltimore: Johns Hopkins University Press. p223–248.

Alemseged Z, Wynn JG, Kimbel WH, Reed D, Geraads D, Bobe R 2005. A new hominin from the Basal Member of the Hadar Formation, Dikika, Ethiopia, and its geological context. J Hum Evol 49:499–514.

Bailey SE. 2004. A morphometric analysis of maxillary molar crown of Middle-Late Pleistocene hominins. J Hum Evol 47:183–198.

Bailey SE, Pilbrow VC, Wood BA. 2004. Interobserver error involved in independent attempts to measure cusp base areas of Pan M1s. J Anat 205:323–31.

Beynon AD, Dean MC, Reid DJ. 1991. On thick and thin enamel in hominoids. Am J Phys Anthropol 86:295–310.

Beynon AD, Wood B. 1987. Patterns and rates of enamel growth on the molar teeth of early hominids. Nature 326:493–496.

Boyde A. 1989. Enamel. In: Berkovitz BKB, Boyde A, Frank RM, Hohling HJ, Moxham BJ, Nalbandian J, Tonge CH, editors. Teeth. Handbook of microscopic anatomy , Vol.6. Berlin: Springer. p309–473.

Bromage TG. 1987. The biological and chronological maturation of early hominids. J Hum Evol 16:257–272.

Bromage TG, Dean MC. 1985. Re-evaluation of the age at death of immature fossil Hominids. Nature 317:525–527.

Bromage TG, Schrenk F, Zonnenveld FW. 1995. Palaeoanthropology of the Malawi Rift: an early hominid mandible from the Chiwondo Beds, northern Malawi. J Hum Evol 28:71–108.

Dean MC. 2006. Tooth microstructure tracks the pace of human life-history evolution. Proc R Soc B 273:2799–2808.

Dean MC. 2010. Retrieving chronological age from dental remains of early fossil hominins to reconstruct human growth in the past. Philos Trans R Soc Lond B Biol Sci 365:3397–410.

Dean C, Leakey MG, Reid D, Schrenk F, Schwartz GT, Stringer C, Walker A. 2001. Growth processes in teeth distinguish modern humans from *Homo erectus* and earlier hominins. Nature 414:628–631.

Grine FE, Smith HF, Heesy CP, Smith EJ. 2009. Phenetic affinities of Plio-Pleistocene *Homo* fossils from South Africa: molar cusp proportions. In: Grine FE, Fleagle JG, Leakey RE, editors. The first humans: origin and evolution of the genus *Homo*. Dordrecht: Springer. p49–64.

Grine FE, Ungar PS, Teaford MF. 2006. Was the Early Pliocene hominin '*Australopithecus*' *anamensis* a hard object feeder? S Afr J Sci 102:301–310.

Haile-Selassie Y. 2010. Phylogeny of early *Australopithecus*: new fossil evidence from the Woranso-Mille (central Afar, Ethiopia). Philos Trans R Soc Lond B Biol Sci 365:3323–3331.

Haile-Selassie Y, Saylor BZ, Deino A, Alene M, Latimer BM. 2010. New hominid fossils from Woranso-Mille (Central Afar, Ethiopia) and taxonomy of early *Australopithecus*. Am J Phys Anthropol 141:406–417.

Johanson DC, White TD, Coppens Y. 1982 Dental remains from the hadar formation. Ethiopia: 1974–1977 collections. Am J Phys Anthropol 57:545–603.

Kelley J, Smith TM. 2003. Age at first molar emergence in early Miocene *Afropithecus turkenensis* and life history evolution in the Hominoidea. J Hum Evol 44:307–329.

Kimbel WH, Delezene LK. 2009. "Lucy" redux: a review of research on *Australopithecus afarensis*. Am J Phys Anthropol 140:2–48.

Kimbel WH, Lockwood CA, Ward CV, Leakey MG, Rak Y, Johanson DC. 2006. Was *Australopithecus anamensis* ancestral to *A. afarensis*? A case of anagenesis in the hominin fossil record. J Hum Evol 51:134–152.

Lacruz RS, Dean MC, Ramirez Rozzi FV, Bromage TG. 2008. Megadontia, patterns of enamel secretion, and striae periodicity in Plio-Pleistocene fossil hominins. J Anat 213:148–158.

Lacruz RS, Ramirez Rozzi FV. 2010. Molar crown development in *Australopithecus afarensis*. J Hum Evol 58:201–206.

Lacruz RS, Ramirez Rozzi F, Bromage TG. 2005. Linear enamel hypoplasia, weaning, and age at death in the Taung child. S Afr J Sci 101:567–569.

Lacruz RS, Ramirez Rozzi FV, Bromage TG. 2006. Variation in enamel development of South African fossil hominids. J Hum Evol 51:580–590.

Leakey MG, Feibel CS, McDougall I, Walker A. 1995. New four million-year-old hominid species from Kanapoi and Allia Bay. Nature 376:565–571.

Lucas PW, Corlett RT, Luke DA. 1986. Postcanine tooth size and diet in anthropoids. Z Morphol Anthropol 76:253–276.

Macho GA. 2001. Primate crown formation time and life history evolution revisited. Am J Primatol 55:189–201.

Mahoney P. 2008. Intraspecific variation in M1 enamel development in modern humans: implications for human evolution. J Hum Evol 55:131–147.

Moggi-Cecci J, Boccone S. 2007. Maxillary molar cusp morphology of South African australopithecines. In: Bailey S, Hublin J-J, editors. Dental perspectives in human evolution: state of the art research in dental paleoanthropology. Dordrecht: Springer. p53–64.

Quam R, Bailey S, Wood BA. 2009. Evolution of M^1 crown size and cusp proportions in the genus *Homo*. J Anat 214:655–670.

Ramirez Rozzi F. 1993. Tooth development in East African *Paranthropus*. J Hum Evol 24:429–454.

Reid DJ, Dean MC. 2006. Variation in modern human enamel formation times. J Hum Evol 50:329–346.

Reid DJ, Schwartz GT, Dean C. Chandrasekera MS. 1998. A histological reconstruction of dental development in the common chimpanzee, *Pan troglodytes*. J Hum Evol 35:427–448.

Schwartz GT, Mahoney P, Godfrey LR, Cuozzo FP, Jungers WL, Randria GF. 2005. Dental development in *Megaladapis edwardsi* (Primates, Lemuriformes): implications for understanding life history variation in subfossil lemurs. J Hum Evol 49:702–721.

Smith BH. 1991. Dental development and the evolution of life history in the Hominidae. Am J Phys Anthropol 86:157–174.

Smith TM, Reid DJ, Dean MC, Olejniczak AK, Martin LB. 2007. Molar development in the common chimpanzee (*Pan troglodytes*). J Hum Evol 52:201–216.

Strait DS, Grine FE. 2004. Inferring hominoid and early hominid phylogeny using craniodental characters: the role of fossil taxa. J Hum Evol 47:399–452.

Suwa G. 1988. Evolution of the "robust" australopithecines in the Omo succession: evidence from mandibular premolar morphology. In: Grine FE, editor. Evolutionary history of the "robust" australopithecines. New York: Aldine de Gruyter. p 199–222.

Suwa G, Wood BA, White TD. 1994. Further analysis of mandibular molar crown and cusp areas in Pliocene and early Pleistocene hominids. Am J Phys Anthropol 93:407–426.

Suwa G, White TD, Howell FC. 1996. Mandibular postcanine dentition from the Shungura Formation, Ethiopia: crown morphology, taxonomic allocations, and Plio-Pleistocene hominid evolution. Am J Phys Anthropol 101:247–282.

Suwa G, Kono RT, Simpson SW, Asfaw B, Lovejoy CO, White TD. 2009. Palaeobiological implications of the *Ardipithecus ramidus* dentition. Science 326:94–99 (and Supporting Online Material).

Teaford MF, Ungar PS. 2000. Diet and the evolution of the earliest human ancestors. Proc Natl Acad Sci USA 97:13506–13511.

Ungar PS, Scott RS, Grine FE, Teaford MF. 2010. Molar microwear textures and the diets of *Australopithecus anamensis* and *Australopithecus afarensis*. Philos Trans R Soc Lond B Biol Sci 365:3345–3354.

Ward CV, Leakey MG, Walker A. 2001. Morphology of *Australopithecus anamensis* from Kanapoi and Allia Bay, Kenya. J Hum Evol 41:255–368.

Ward CV, Plavcan JM, Manthi FK. 2010. Anterior dental evolution in the *Australopithecus anamensis-afarensis* lineage. Philos Trans R Soc Lond B Biol Sci 365:3333–3344.

White TD. 1977. New fossil hominids from Laetolil, Tanzania. Am J Phys Anthropol 46:197–230.

White TD, Suwa G, Simpson S, Asfaw B. 2000. Jaws and teeth of *Australopithecus afarensis* from Maka, Middle Awash, Ethiopia. Am J Phys Anthropol 111:45–68.

White TD, WoldeGabriel G, Asfaw B, Ambrose S, Beyene Y, Bernor RL, Boisserie JR, Currie B, Gilbert H, Haile-Selassie Y, Hart WK, Hlusko LJ, Howell FC, Kono RT, Lehmann T, Louchart A, Lovejoy CO, Renne PR, Saegusa H, Vrba ES, Wesselman H, Suwa G. 2006. Asa Issie, Aramis and the origin of *Australopithecus*. Nature 440:883–889.

White TD, Asfaw B, Beyene Y, Haile-Selassie Y, Lovejoy CO, Suwa G, WoldeGabriel G. 2009. *Ardipithecus ramidus* and the paleobiology of early hominids. Science 326:75–86.

Wood BA. 2010. Reconstructing human evolution: achievements, challenges, and opportunities. Proc Nat Acad Sci USA 107:8902–8909.

Wood BA, Abbott SA, Graham SH. 1983. Analysis of the dental morphology of Plio-Pleistocene hominids. II. Mandibular molars: study of cusp areas, fissure pattern and cross sectional shape of the crown. J Anat 137:287–314.

APPENDIX: LIST OF SPECIMENS USED IN THIS STUDY

	Au. anamensis		*Au. afarensis*	
	Specimen	Locality	Specimen	Locality
M1	KNM-KP 29286	Kanapoi	AL128–23	Hadar
	KNM-KP 29281	Kanapoi	AL145–35	Hadar
	KNM-KP 34725	Kanapoi	AL200–1b	Hadar
	KNM-KP 31712	Kanapoi	AL266–1	Hadar
	KNM-KP 30500	Kanapoi	AL288–1	Hadar
	KNM-ER 30201	Allia Bay	AL333–74	Hadar
	KNM-ER 20422	Allia Bay	AL333w-12	Hadar
			AL333w-1	Hadar
			AL333w-60	Hadar
			LH.2	Laetoli
			LH.3	Laetoli
			LH. 4	Laetoli
			LH.16	Laetoli
M2	KNM-KP 29286	Kanapoi	AL128–23	Hadar
	KNM-KP 34725	Kanapoi	AL145–35	Hadar
	KNM-KP 29287	Kanapoi	AL188–1	Hadar
	KNM-KP 30500	Kanapoi	AL198–1	Hadar
	KNM-KP 29281	Kanapoi	AL207–13	Hadar
	KNM-ER 35233	Allia Bay	AL266–1	Hadar
			AL277–1	Hadar
			AL288–1	Hadar
			AL333w-1	Hadar
			AL333w-27	Hadar
			AL333w-57	Hadar
			AL333w-59	Hadar
			AL333w-60	Hadar
			AL400	Hadar
			LH.4	Laetoli
			LH.19	Laetoli
			LH.23	Laetoli
			MAK1–2	Maka
			MAK1–3	Maka
			MAK1–12	Maka
M3	KNM-KP 29286	Kanapoi	AL188–1	Hadar
	KNM-KP 30500	Kanapoi	AL198–1	Hadar
	KNM-KP 29281	Kanapoi	AL266–1	Hadar
	KNM-ER 20428	Allia Bay	AL288–1	Hadar
			AL333–59	Hadar
			AL333–74	Hadar
			AL333w-57	Hadar
			AL333w-59	Hadar
			AL333w-32/60	Hadar
			AL400–1	Hadar
			AL366–1	Hadar
			LH.4	Laetoli
			LH.15	Laetoli
			MAK1–2	Maka
			MAK1–12	Maka